BORING SPONGE INFESTATION ON THE MUSSEL *PERNA INDICA* KURIAKOSE AND NAIR 1976 FROM THE SOUTHWEST COAST OF INDIA

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Certified that the thesis entitled " **BORING SPONGE INFESTATION ON THE MUSSEL *PERNA INDICA*. KURIAKOSE AND NAIR FROM THE SOUTHWEST COAST OF INDIA** " is a record of independent bonafide research work carried out by **Shri. P. Sunil kumar** during the period of study from September, 1997 to September, 2000 under our supervision and guidance for the degree of **Doctor of philosophy in Fish and Fisheries Science (Mariculture)** and that the thesis has not previously formed the basis for the award of any degree, diploma, associateship, fellowship or any other similar title.

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DECLARATION

I hereby declare that this thesis entitled "**BORING SPONGE INFESTATION ON THE MUSSEL *PERNA INDICA* KURIAKOSE AND NAIR 1976 FROM THE SOUTHWEST COAST OF INDIA** " is an authentic record of the work done by me and that no part thereof has been presented for the award of any degree, diploma, associateship, fellowship or any other similar title.

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सारांश

भारत के दक्षिण - पश्चिम तट के छः चुने गए केंद्रों में वर्ष 1998-2000 की अवधि के दौरान भुरा शंबु (*पेर्ना इंडिका* कुरियाकोस और नायर,1976) के प्राकृतिक तथा संवर्धित स्टॉक में वेधन स्पंजों के ग्रसन के तरीके पर अध्ययन किया गया और इसके प्रमुख परिणाम इस शोध प्रबंध में प्रस्तुत किए जाते हैं. उपर्युक्त अवधि के दौरान वेधन स्पंजों की कुल नौ जातियाँ शंबु संस्तरों पर ग्रसन करते हुए देखा गया और कवचों में देखी गई इनकी असंख्य प्रचुरता के अनुसार ये नौ जातियाँ हैं : *क्लयोना वास्टिफिका, क्लयोना लोबाटा, क्लयोना मारगरिटिफेरा, क्लयोना सेलाटा, क्लयोना कारपेन्टेरी, थूसा हानकोकी थूसा, थूसा अरमाटा, अका मैन्यूटा और अलेक्टोना मिल्लारी. क्लयोना वास्टिफिका* पृथुलवणी होने की वजह से इस तट के ज्वारनदमुखों में व्यापक है.

उपर्युक्त नौ जातियों में से *सी. मारगरिटिफेरा* और *सी. लोबाटा*, जो शंबुओं के दो भयानक नाशक जीव हैं, वर्ष 1980 के दौरान विषिंजम क्षेत्र (संवर्धन राफ्टों में) तक प्रवास किया है और तब से लेकर ये दक्षिण-पश्चिम तट और मान्नार खाडी के प्राकृतिक शंबु स्टॉकों तक प्रवास करने लगे और ग्रसन तरीका, जाति मिश्रण आदि में विचारणीय तेज़ी लाने लगे. वर्तमान अध्ययन जो विषिंजम में इनकी पहली उपस्थिति के 20 वर्षों के बाद किया जाता है, इन जीवों के परंपरागत जातियों के साथ परस्पर प्रभाव जैसे गतिविधियों का अनुपरीक्षण व्यक्त करता है. वर्तमान अध्ययन में, अटलान्टो - मेडिटरेनियन क्षेत्र में पाए जाने वाले प्रवाल वेधन स्पंज (अलेक्टोना मिल्लारी) को उपर्युक्त छः केंद्रों में से एक केंद्र में दिखाया पडा. शंबुओं से, प्रवालों तथा अन्य कवचों के नाशकारी दो जातियों को भी प्राप्त हुआ. विभिन्न केंद्रों से संग्रहित नमूनों को उपयुक्त करके वेधनकारी और अवेधनकारी शंबुओं की लंबाई आवृत्ति , जाति मिश्रण, ग्रसन तरीका आदि का विवरण रिकार्ड करके उनके माहिक और वार्षिक आकलन किया गया. शंबुओं में स्पंजों के ग्रसन से होने वाले 6 रोगों में फफोला रोग प्रमुख (46%) देखा गया. इस रोग से मुक्ताभ वस्तुओं का अधिक रूप से नाश हो जाता है और मुक्ता शुक्ति में यह रोग मोती उत्पादन में प्रतिकूल प्रभाव डालता है. अध्ययन के एक भाग के रूप में आधुनिक प्रौद्योगिकियों द्वारा शंबु के मृदु भाग में हुए रोगजनक परिवर्तन ट्रेस किया गया. संग्रहित जातियों का विवरण अनुयोज्य व्याख्या एवं निर्वचनों के साथ किया गया.

ABSTRACT

The pattern of infestation of boring sponges on natural as well as tended stocks of brown mussel, *Perna indica* from six selected centres along the southwest coast of India during 1998-2000 period was studied and the salient findings emerged are presented in this thesis. A total of nine species of boring sponges was found to infest the mussel beds during the above period and the various species as per their numerical abundance in total shells examined are as follows: *Cliona vastifica*, *Cliona lobata*, *Cliona margaritifera*, *Cliona celata*, *Cliona carpenteri*, *Thoosa hancocki*, *Thoosa armata*, *Aka minuta* and *Alectona millari*. *Cliona vastifica*, since euryhaline, is distributed in the estuaries of this coast. Of the above nine species, *C. margaritifera* and *C. lobata*, two dreadful pests, elsewhere, have migrated to Vizhinjam area (culture rafts) around 1980 and since then have migrated to wild molluscan stocks along the southwest coast and thence to Gulf of Mannar causing considerable hike in the infestation pattern, species composition etc. The present study, made after 20 years from their first appearance at Vizhinjam, hence may be taken as a follow up in tracing their activities including their interaction with conventional species. During the present study the invasion of another common coral boring sponge of the Atlanto-Mediterranean zone (*Alectona millari*) could be detected from one of the above six centres. Two species common to coral/ other shells could also be collected from mussels as pests. Utilising the samples collected regularly from different centres, details on length frequency distribution of both bored and unbored mussels, species composition, and pattern of incidence etc. were recorded and their monthly and yearly projections were made. Of six diseases caused in mussels due to sponge infestation blister formation was predominant (46.3 %). Pathological changes effected in the soft part of mussel were traced out with electron microscopic techniques. Species collected have been described with suitable illustrations and a key also is provided.

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INTRODUCTION

Aquaculture of molluscs has been growing at an average annual rate of 11 % over the past decade, compared to that of livestock meat and capture fisheries which showed only 3.1 % and 1.6 % respectively. Main productions are oysters, contributing to 36% of the total molluscs, clams 23 %, scallops 15 %, and mussels 13 % (FAO, 1997).

Disease outbreaks are increasingly recognized as a significant constraint to aquaculture production and trade, affecting both the economic development and socio-economic revenue of many countries. Molluscan farming in many countries faces serious disease problems resulting in significant production losses. One of the very few ways to reduce the impact of such pathogens and borers on commercially exploited molluscs, is likely to establish effective programmes to prevent the transfer of infested stocks. Demospongiae of the family Clionidae are known for their capacity to burrow or bore into calcareous objects including the shells of dead and living molluscs. Sponges with the ability to etch into and live in calcareous objects have been identified within the families Adociidae, Clionidae, Spirastrellidae, Jaspiidae and Halinidae of the Class Demospongiae of the Phylum Porifera. The genus *Cliona* with about 65 species is the best known as borers. The members of the family Clionidae are important as pests in molluscan aquaculture (Alagarswami and Chellam 1976; Thomas, 1979).

Molluscan culture is gaining momentum in several parts of India. The transfer of technology programmes conducted by Central Marine Fisheries Research Institute, Cochin has given new impetus to mussel culture activities especially in the estuarine areas of Kerala. Knowledge of the boring and fouling organisms is inevitable from molluscan culture point of view. The success of any culture system depends on several factors like the quality of seed used, growth rate of the target group etc. Apart from external damage, boring organisms may reduce growth and affect nutritional quality of the cultured groups. The foulers as well as borers directly control these factors. About 12 major taxa of marine algae and invertebrates have been included

under the category of boring organisms. Typical examples are clionid sponges, polychaetes, boring molluscs, sipunculids and algae.

Boring sponges have been known to cause considerable damage to the commercial shell-fish stocks since they were discovered at the beginning of the 1800's in French oyster beds, where they were reported to cause 'spice bread disease' (Thomas, 1981). In recent years clionids have been documented from Black Sea oysters (Krakatitsa and Kaminskaya, 1979), edible oyster culture in the Indian seas (Thomas, 1979 B; 1983), pearl oyster and mussel culture farms in Japan and India (Alagarswami and Chellam, 1978; Thomas *et al.*, 1983).

Boring sponges of the genus *Cliona* may be found in three developmental stages: burrowing into calcareous material (α stage); completely encrusting the original objects they have eroded (β stage); and massive free-living, leaving no signs of the original excavated material (γ stage). In tropical conditions, normally only the first stage is reached (Rosell and Uriz, 1994). After the death of the living sponge, cavities and sometime loose spicules may remain for longer time (Rosell, 1994).

Field identification of *Cliona* species is difficult since the boring pattern of all species is more or less the same. Hence microscopic examination of spicules is highly essential for specific identification. But sponges are unique in the respect that the cavities produced inside the substratum follow a definite pattern: " chamber and canal pattern. "The branch of sponge running through the inner layers of the shell expands to form a chamber after a short distance ("sponge mass") and from this "mass" another branch is formed, and this will form another chamber. This pattern is subject to considerable variation based on the substratum and the stage of boring. In mussels and pearl oysters infestation starts at the umbo region, whereas in edible oysters, the attached valve is the prime target. In gastropods like *Xancus pyrum* the spire area is initially affected.

The relationship between boring sponges and their host, (shell/coral) has been termed "parasitism" for a long period. In this association sponge is benefited by getting a substratum of calcium carbonate to grow and proliferate while the host (mollusc/ coral) is affected adversely. There is no trophic relation between sponge and its host. This sort of a relationship is now termed 'ammensalism' (Krakatitsa and Kaminskaya, 1979).

In India two species of mussels, green mussel (*Perna viridis*, Linnaeus) and brown mussel *(Perna indica*, Kuriakose and Nair) are exploited for culinary purpose. The brown mussel, *Perna indica*, has a limited distribution along the Indian coast: from Kollam to Kanyakumari along the west coast and from Leepuram to Chinnamuttom along the east coast in a total area of 50 square kilometers. The major landing centres along the west coast are Varkala, Kovalam, Vizhinjam, Mulloor, Poovar, Enayam, Colachel, Kadiyapatnam, Muttom, and Chinnamuttom. Mussel fishing is done throughout the year, except during monsoon season. Peak landings have been reported in the months of September and December. It is interesting to note in this context that a species of boring sponge, *C. vastifica*, is capable of invading low salinity areas due to its euryhaline nature and hence the molluscan culture farms along the Indian coast are under severe threat of *C. vastifica* attack (Thomas, 1975). Bivalve mariculture has the potential for supplying the much needed animal protein, generating employment opportunities in coastal areas and for meeting the growing demand in international market. Since the growth of farmed molluscs is considerably influenced by the attack of boring organisms and foulers, a detailed investigation on the taxonomy, distribution, migration pattern, control measures etc. of these organisms is essential.

The thesis consists of six chapters. The first and second chapters are on the pattern and mechanism of boring, and the third one is on taxonomy of mussel boring sponges. The fourth chapter deals with species composition, distribution, abundance and biodiversity of boring sponges along the study area. The fifth chapter is on boring sponge infestation in molluscan culture systems Histopathological and ultrastructure

aspects are dealt with in the succeeding chapter. Each chapter consists of introduction, review of literature, material and methods, results and discussion. List of references is included at the end.

Considering the importance of borers in mussel culture, this study was taken up to discuss the effect of *Cliona* infestation on *Perna indica* along the southwest coast of India. The main aspects studied are the nature and intensity of sponge infestation in different size groups, the effect of infestation on the anatomy of mantle tissue, quality of mussel meat based on biochemical assays, and the methods to eradicate sponge infestation on *Perna indica* culture systems.

The objectives of the present study are:

1. To identify the species of boring sponges infesting the molluscan shells of the southwest coast with particular reference to the brown mussel population.
2. To estimate the extent of incidence, species composition and competition for dominance of boring sponges.
3. To evaluate the extent of damage caused to the shell and associated pathological symptoms.
4. To compare boring sponge infestation on farmed stocks with that in the natural beds.
5. To study tissue-level changes in mantle and adductor muscle.
6. To study the ultrastructure of uninfested and sponge-infested mantle tissue.

1. PATTERN OF BORING

1. INTRODUCTION

The primary aim of the present study was to make a detailed survey of the boring species of sponges infesting the brown mussel; *Perna indica*, Kuriakose and Nair, 1976 distributed along the southwest coast of India. Brown mussel samples, hence, were collected from some selected centers (Map 1) at regular intervals and bored shells present in these samples were utilized for the present study. A total of nine species of boring sponges could be collected from the above samples, and a list is given in Chapter 3. But considering the richness of other molluscan species, both gregarious and otherwise, along the southwest coast from Kollam (= Quilon) to Cape Comorin (= Kanyakumari) and the availability of bored shells in plenty, it was decided to make a comprehensive study of various species of boring sponges infesting the molluscs in general along the above area.

2. MATERIAL AND METHODS

A. Collection centres

Specimens for the present study were collected from six centres along the southwest coast of India (Map 1). The stations from north to south are: 1. Vizhinjam; 2. Mulloor; 3. Enayam; 4. Colachel; 5. Kadiyapatnam; 6. Cape Comorin

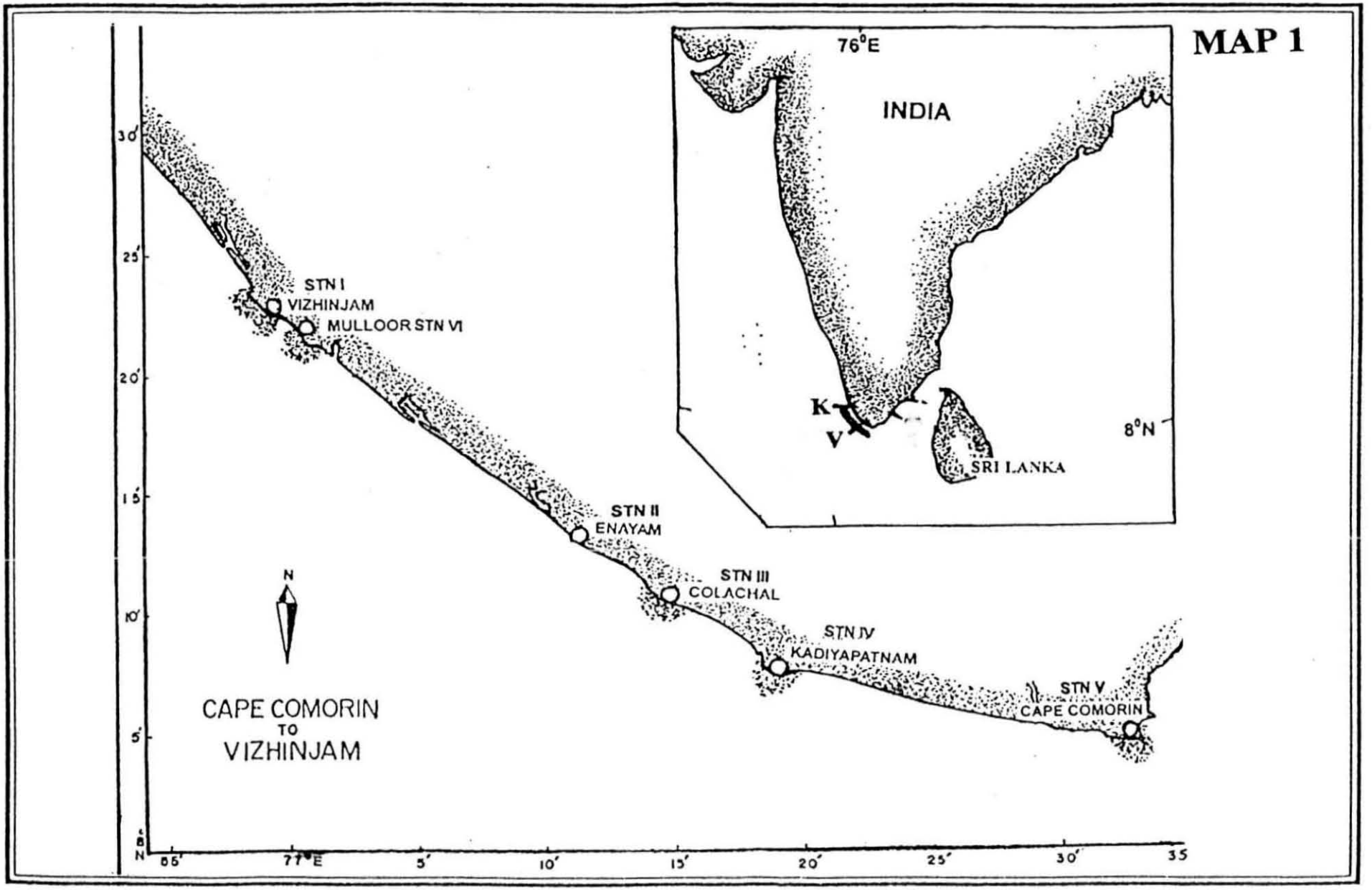
B. Collection methods

Samples were collected from the above said centres once/twice a month. The mussels brought ashore regularly by the mussel pickers (skin-divers) in the morning hours were mainly utilised in the present study. These collections were supplemented with those collected from the intertidal areas by the worker himself.

Map 1. Map showing the sampling Stations i-vi

K. Kollam (Quilon)- Estuarine centre

V. Vizhinjam- Raft cultured brown mussel


MAP 1
76°E
INDIA
K
V
8°N
SRI LANKA
STN I
VIZHINJAM
MULLOOR STN VI
STN II
ENAYAM
STN III
COLACHAL
STN IV
KADIYAPATNAM
STN V
CAPE COMORIN
N
CAPE COMORIN
TO
VIZHINJAM
30'
25'
20'
15'
10'
5'
8°N
55'
77°E
5'
10'
15'
20'
25'
30'
35

A minimum of 100 shells was randomly sampled each time and in centres where more than one mussel picking units are involved, the samples were collected from each unit proportionately to make the total number 100. While collecting the specimens special care was taken to collect all size groups present in the landings.

C. Preliminary examination

The specimens, soon after collection, were examined for the colour of the sponge, size of papillary projections etc. and then they were cleaned from biofoulants in the field itself. These were then washed thoroughly in seawater and transported to the laboratory, partly soaked in seawater.

D. Laboratory examination

In the laboratory the total length of each mussel was recorded and then the bored and unbored shells were separated. In shells where boring was in its initial stages a lens (10 x) was used to locate the pores which were so minute and scanty in distribution. The data, thus collected, were used to find out the incidence (infestation/100 shells) of boring sponges in that particular bed, as also the size frequency distribution of the bored fraction in relation to the entire lot. Bored shells separated form the whole lot was then examined individually to see whether the right, left or both the valves are infested with sponge (Fig. 1). The area and extend of infestation (by noting the pores through which the excurrent and incurrent papillae project out) were then recorded. When the umbo portion alone is infested it is often given as 1/8 and likewise 1/2, 3/4 and finally full when the shell is completely beset with openings bearing both incurrent and excurrent papillae at the outer surface. The above examination was done with the help of a hand lens of 10 x magnification.

The valves of the shell were separated and then the soft parts attached to the inner side of the shell were also removed carefully. A detailed examination

of the inner surface of the shell is necessary to assess the damage done to the live mollusc by boring sponges. Blisters and openings may be seen in the inner aspect of the shell in many cases; blisters may be with black pigment at their summit or with openings for the protrusion of sponge papillae into the cavity between the shell and mantle tissue of the living shell for the intake and expulsion of water. Other aspects like roughness of the interior of the shell, pigmentation, nacreous layer erosion etc. were also recorded.

When the sponge papillae project out and touch the mantle epithelium in living condition, several local reactions and pathological manifestations are bound to occur on the soft tissue of the mantle. The mantle epithelium may develop discoloration due to the papillar contacts and hence such tissues, when observed, were preserved in Bouins fixative and Osmium tetroxide (OsO_4) for further studies.

E. Spicule preparation

The method suggested by Old (1941) was followed in general with some modifications. For extracting the spicules a bit of shell infested by sponge was placed in a test tube and concentrated nitric acid was poured into it and then heated over a low flame till the calcareous shell is completely dissolved. The test tube is then filled with water; stirred thoroughly and kept aside to allow the spicules to settle down. The supernatant water was then poured out gently and replaced with clean water. This process is continued for about 3-4 times.

The residue thus obtained is a "spicule concentrate" and is almost free from nitric acid particles. One or two drops of the same were transferred to a micro slide by means of a fine dropper, spread it evenly and then mounted with water as the medium. Several drops were examined under different combination of lenses and necessary drawings were prepared using a camera lucida. Various measurements (length, width etc.) were taken with the help of a calibrated ocular micrometer. The use

of 100 measurements for each spicule category was avoided and instead 10 measurements/ category was adopted following Bakus (1966). Spicule measurements are expressed in mm and a uniform pattern, lower limit and upper limit is followed uniformly for each spicule category. For making permanent slides, spicules were washed thoroughly in distilled water and then in absolute alcohol. One or two drops of "spicule concentrate" were taken on a micro slide and after spreading evenly over the slide, it was allowed to dry up in room temperature. After the evaporation of alcohol, a few drops of xylol was poured over the spicules and again allowed to evaporate fully. For making permanent slides EUPARAL (Product No. 928375) was used as the medium.

In the initial stages the sponge infestation is confined to the thickest parts of shell. But in advanced stages, the ramifications of sponge may reach upto the margin of the shell. The pores produced at the surface of the shell by various species of sponges are of different size and pattern and hence various species of *Cliona* are very difficult to be identified in the field by naked eye. Plate 1 shows the openings at the outer surface of the shell by *Cliona lobata*.

F. Examination of sections of the shells

To study the proliferation of boring sponge inside the shell, the extend of damage done to the shell, the pathological manifestations akin to sponge boring etc. it is necessary to take sections of the shell, both vertical and horizontal. Since it is difficult to take thin sections of the shell in any desired plane, thick sections were made use of in most cases. Sections, thus prepared in any desired plane, were mounted on a rigid support (cork/thermocoal) and examined under low power (2.5 × 6) of the microscope. The details of chambers, interchamberal canals, surface pores through which incurrent and excurrent papillae protrude out, blisters produced at the inner side of the shell, etc, were studied in this way and necessary sketches were prepared using different combination of lenses (2 × 6x; 6 × 45x etc.). All measurements were taken with

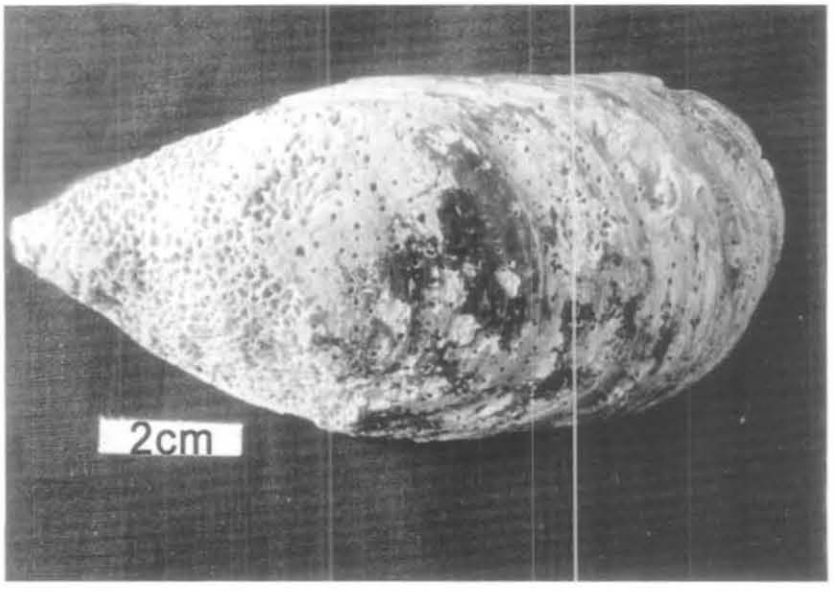


Plate 1. *Perna indica* shell (left valve) infested by boring sponge *Cliona lobata* (Scale=2cm)

the help of calibrated occular micrometer and then they were converted to millimeter with the constant arrived at for the different combination of lenses. Right and left valves of *Perna indica* various measurements adopted etc. are shown in Fig. 1.

The umbo part of the shell is the thickest and this region is usually infested first. In a vertical section of the umbo the chambers formed inside by sponge may extend from the periostracum to the nacreous layer vertically and the chambers formed may be even in different tiers (Fig. 2). But in thinner parts of the shell the chambers made by sponges may be in a single tier (Fig. 3).

A. Horizontal section

To study the lateral expanse and nature of chambers formed inside the shell by boring sponges horizontal sections were taken as shown in Fig. 2 and 3. It is very easy to make horizontal sections since the shell is made up of three different layers, periostracum, prismatic layer and nacreous layer (Fig. 4) and this can be done easily with the help of a razor blade or fine needle in the desired plane by applying slight force.

1. in the thickest part (Fig. 2), of umbo proper, it is necessary to take horizontal sections through ULC and LLC, ie. through the upper and lower layer of chambers and
2. in the thinner parts since there is only one layer of chambers the sections may be preferred along the middle of the layer of chambers (LC).

Details and angle of examination

US - The upper surface

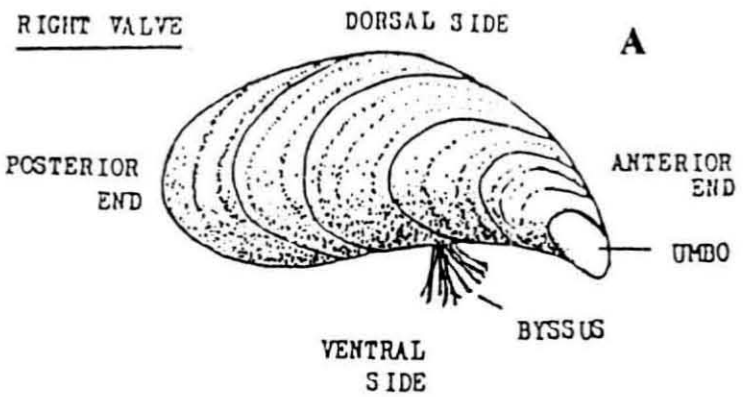
The upper surface of the shell may be examined by keeping the shell horizontally with convex part directed upwards and viewing the surface as

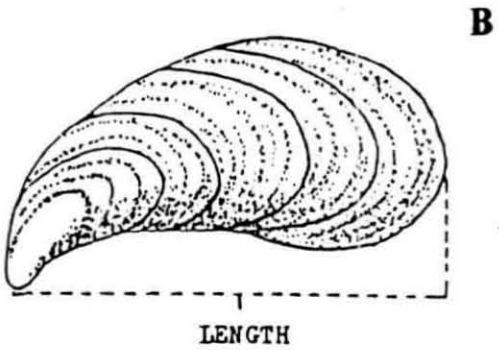
Fig. 1 Parts of brown mussel shell

A. Anterior and posterior ends, umbo region and adductor scar of left valve
B. Length of mussel is recorded as per the specifications shown.
C. Left valve, inner view, showing the adductor scar

FIGURE 1

BROWN MUSSEL - PERNA INDICA





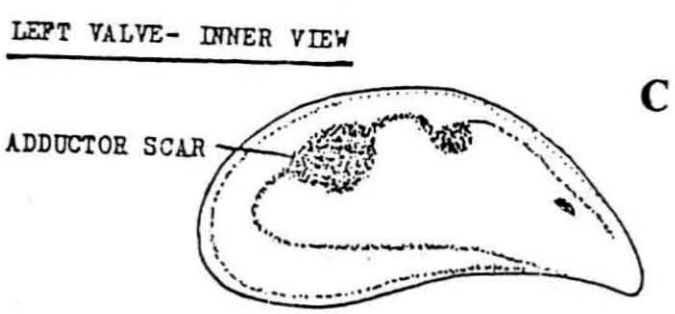


***Fig.* 2 Vertical and horizontal section of the shell to be prepared and details to be studied (brown mussel shell; infested with *Cliona margaritifera*; umbo part)**

A. Vertical section (V.S)

Vertical section through the thickest part of a shell (umbo part showing the proliferation of sponge (stippled areas) inside the shell (the original shell is shown by unstippled areas). Chambers and canals (CH and ICC) formed by the sponge completely fill the shell from the upper surface (US) to lower surface (LS). Papillae of sponge open to the exterior through pores both at the upper and lower surfaces alike. The excurrent papillae (EPA) protrude through excurrent pores (EP) and incurrent papillae (IPA) through incurrent pores (IP) at both surfaces alike in living condition.

The mussel, in living condition, takes many precautions to ward off the sponge papillae from touching the soft mantle tissue. Pores made in the inner surface (nacreous layer) are closed by secreting additional nacreous material by the living mussel. But when the mussel become old or weak, the openings made by sponge papillae remain open throughout resulting in perpetual irritation to the mantle surface. This may lead to several pathological symptoms like blister formation (B), pigmentation (P), patchy pigmentation (PPI) etc.

In the section given here the sponge is seen ramifying in the interior of the shell in a "chamber and canal pattern", which is unique for sponges. The chambers are seen in 3-5 tiers filling the prismatic layer (central layer). The periostracum and nacreous layers are damaged only slightly since they are riddled to the minimum by papillae protruding from the chambers (papillar canals), which are narrower. Hence in superficial examination any bored shell may appear intact externally, but the interior may be damaged to the maximum.

As the sponges intensify boring in advanced stages, the chambers inside may get united (united chambers, UC) both vertically and horizontally (See Fig. 7) and finally the entire chambers may get united forming a central tunnel extending in between the periostracum and nacreous layer (See Fig. 9 B).

B. Horizontal section (H.S)

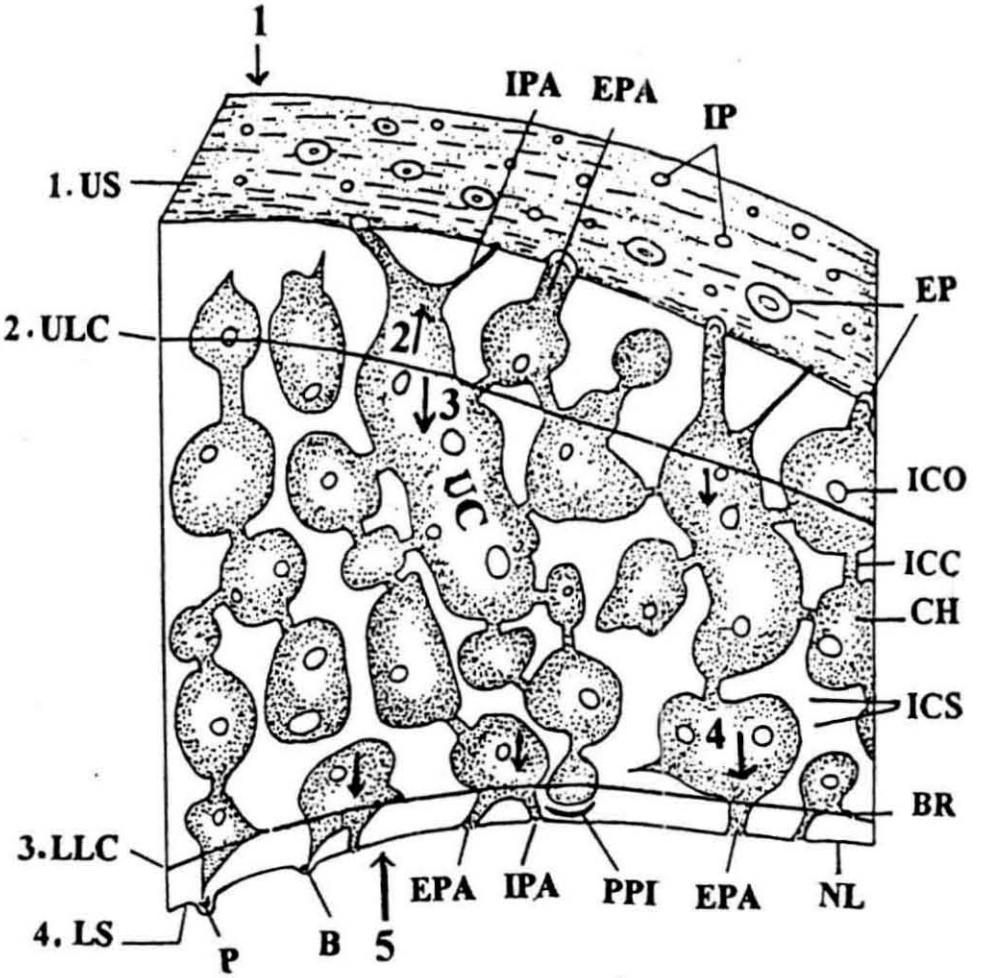
Horizontal section of a shell can be made easily with the help of a razor blade or needle by exerting slight pressure as the shell is made of different layers in its horizontal plane.

After examining the upper and lower surfaces of the shell for pores, blisters etc. in the direction of arrows 1 and 5 respectively, under low power of a microscope (2.5 x 6x) the shell is sliced horizontally at two places: A- At the upper layer of the chambers (ULC) and B- at the lower layer of chambers (LLC). Both the cut

ends at ULC were then examined under low power in the direction of arrow 2 & 3 respectively (See Pl. 2 for a photograph in the direction of arrow 3) for the incurrent and excurrent papillae originating from the chambers and opening at the surface and for the nature, etching pattern, diameter etc. of the chambers. Similarly horizontal sections may be made very close to the nacreous layer where the lower layer of chambers (LLC) are located. This section may be viewed in the direction of the arrow 4 for studying the nature of excurrent and incurrent papillae originating from the lower layer of chambers and openings at the inner side of the nacreous layer (NL). Pigmentation, blister etc. become very clear when viewed in this direction.

B- blister; **BR**- branch of sponge originating from a chamber; **CH**- chamber; **EP**- excurrent pores; **EPA**- excurrent papillae; **IP**- incurrent pores; **ICO**- interchamberal opening; **ICS**- interchamberal septa; **LLC**- lower layer of chambers; **LS**- lower surface of shell; **NL**- nacreous layer; **P**- pigment; **PPI**- plate-like pigment; **US**- upper surface of shell; **UC**- united chambers; **ULC**- upper layer of chambers

FIGURE 2


1
IPA
EPA
IP
1. US
EP
2. ULC
2
3
UC
ICO
ICC
CH
ICS
4
BR
3. LLC
EPA
IPA
PPI
EPA
NL
4. LS
P
B
5

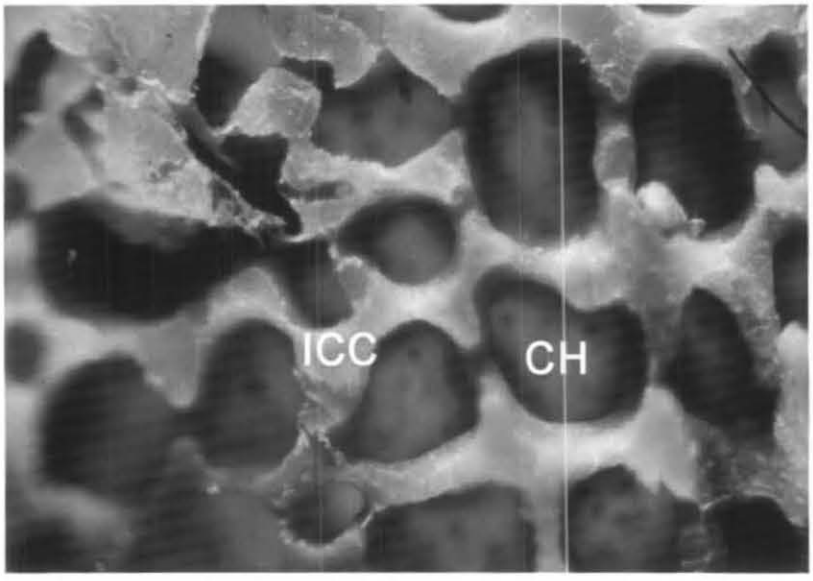


Plate 2. H.S through ULC viewed downwards in the direction of arrow 3 (in Fig. 2) showing magnified view of chambers and inter chamberal canals (CH and ICC)

Fig. 3 Vertical and horizontal sections of a mussel shell bored by sponge (section of thinner part of the shell)

A. Vertical section (V.S)

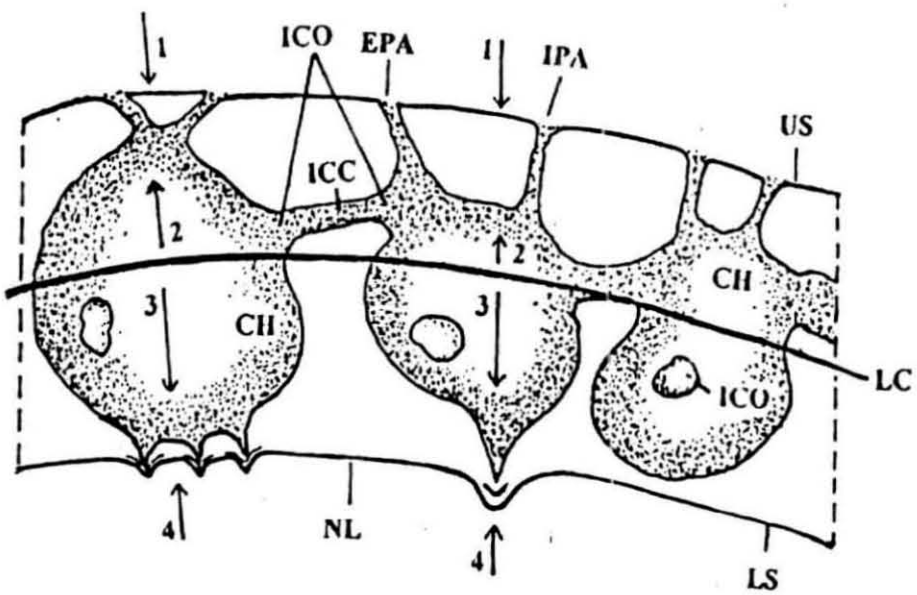
When sponge ramifies through the thinner part (margin of the shell) the chamber and canals are formed in one tier only. The upper surface (US) and lower surface (LS) are studied by placing the shell horizontally on the microscope slide and examining them under low power (2.5 × 6 x) of a microscope in the direction of the arrows 1 and 4. Excurrent papillae (EPA) and incurrent papillae (IPA) as also blisters, pigment etc., could be studied from such vertical sections. The nature of chambers (CH), interchamberal openings (ICO), interchamberal canals (ICC) etc. may also become clearly traceable.

B. Horizontal section (H.S)

Since the sponge proliferates through the interior of the shell and chambers are formed in one tier, horizontal section through the layer of chambers is sufficient to study all the details. Horizontal section can easily be taken by using a razor blade or needle as the shell has a layered structure. The cut made through the layer of chambers (LC), when examined in the direction of arrow 2, will give the details of the upper part of chambers as also the nature of excurrent and incurrent papillae (IPA and EPA) originating from the chambers. The lower section viewed in the direction of arrow 3 give the structure of chambers, etching pattern, incurrent and excurrent papillae originating from the chambers and the way in which they pierce the nacreous layer (NL).

CH- chamber; **EPA**- excurrent papillae; **ICC**- interchamberal canals; **ICO**- interchamberal openings; **IPA**- incurrent papillae; **LC**- layer of chambers; **NL**- nacreous layer; **US**- upper surface

FIGURE 3



shown in Fig. 3 arrow marked 1. This will be helpful in measuring the diameter of excurrent and incurrent pores through which the papillae project out from the surface of the shell.

LS -The lower surface

The lower surface (LS) of the shell may be examined by keeping the shell horizontally with its concave side up and viewing it in the direction of arrow No. 5 in Fig. 2. This is quite helpful in studying the nature of blisters, pustules, openings etc. made inside the shell in advanced stages of infestation.

ULC- The upper layer of chambers.

It is only a horizontal section through the upper layer of chambers, formed by the sponge, inside the shell. This section may be utilised for two purposes, One for examining the chambers and papillar canals originating from the chamber (in the direction of the arrow 2 in Fig. 2) and for studying the diameter, nature etc. of the chambers (in the direction of arrow 3 in Fig. 2).

LLC- The lower layer of chambers

These chambers put forth papillar processes, which pierce the nacreous layer and establish contact with the mantle epithelium of the mussel. This section, when viewed in the direction of the arrow 4, gives a clear picture of the canals leading from the lower part of the chambers to the nacreous layer, pigmentation around such canals, blisters, the canal openings- whether closed or open, etching pattern etc.

Sections preferred in the present study

To study the lateral expanse and the nature of chambers formed inside

the shell by boring sponges, horizontal sections were taken as shown in Figs. 2 and 3. It is easy to make horizontal sections since the shell is made in three different layers, periostracum, prismatic layer and nacreous layer (Fig. 4) and this can be done easily with the help of a razor blade or fine needle in the desired plane by applying slight force.

1. For the thicker part (Fig. 2, umbo proper): it is necessary to take vertical and horizontal sections through ULC and LLC, ie. through the upper and lower layers of chambers and

2. For the thinner parts (Fig. 3): since there is only one layer of chambers, the section may be preferred along the middle layers of the chamber (LC).

"Material and methods" are also provided for each chapter to suit the requirements of the respective study.

Species were identified with the help of Keys and descriptions provided in Bavestrello *et al.*, (1998), Little (1968), Rutzler (1971, 1973, 1974), Rutzler and Stone (1986), Thomas (1972, 1979, 1989) and Schonberg *et al.*, (2000).

3. RESULTS

The free-swimming sponge larva settles down on the surface of the shell (or any calcareous object) and then assumes an encrusting stage and starts boring into the shell by chipping off characteristic particles of the shell from the surface (Fig. 10 D, after Goreau and Hartman, 1963). The larva, after gaining access to the interior of the shell, chips off particles (microchips) liberally and forms an initial chamber (Fig. 5 A, CH-1) inside the shell. Spreading inside the shell is effected in the same way, ie. by chipping off calcareous particles of characteristic size and shape. From the

Fig. 4. Bivalve shell-different layers and mantle epithelium

P- Periostracum
PL- Prismatic layer
NL- Nacreous layer
CM- Cells of mantle

FIGURE 4

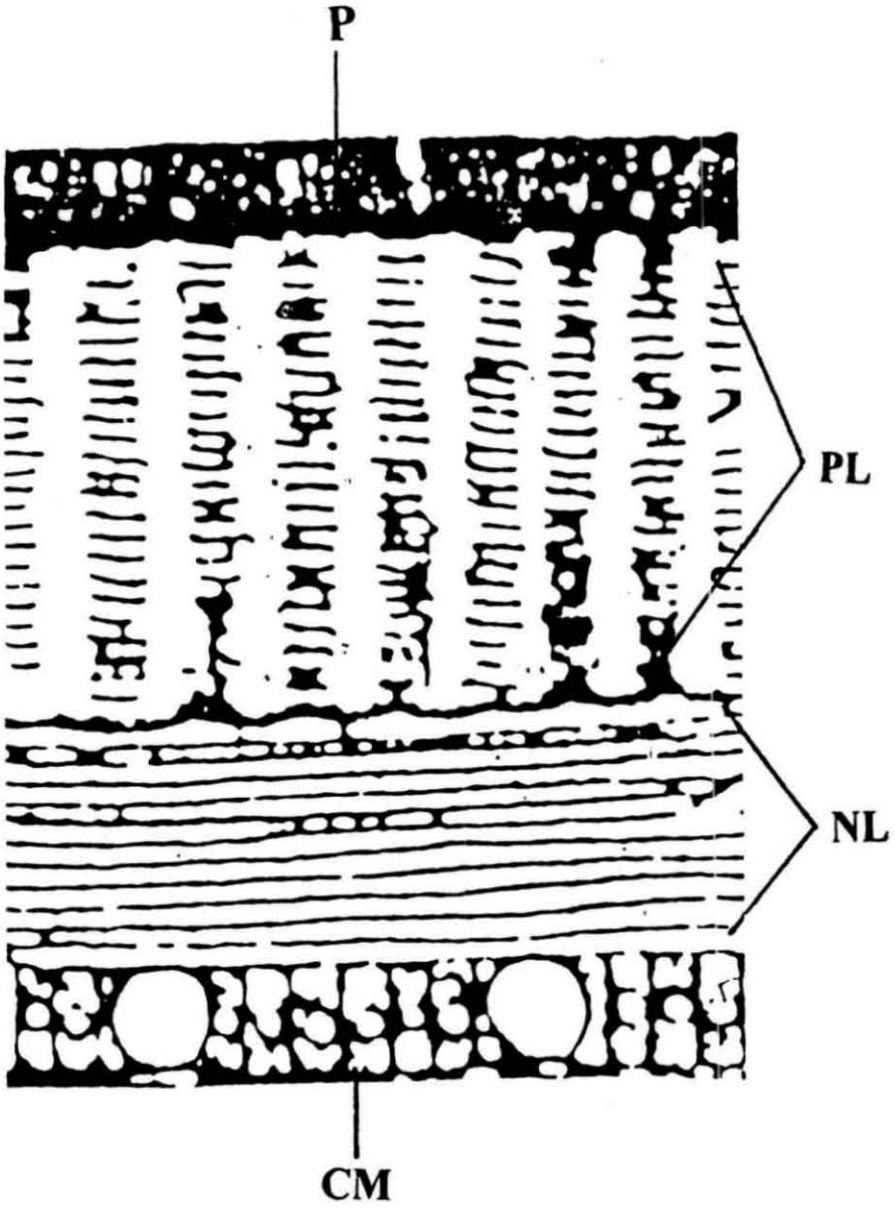


Fig. 5 Formation of the chambers after larval settlement

A. Longitudinal section of shell showing the formation of the initial chamber after the settlement of the sponge larva. Enlargement of the chamber is effected by chipping off calcareous particles from the interior.

B. The boring sponge expands the chamber by growing out at one corner; an initial branch (BR) is thus formed.

C. The above branch grows further by chipping off calcareous matter; a second chamber (CH. 2) is thus formed. From the second chamber branches are formed. Some may be seen directed to the surface of the shell or to the lower surface (nacreous layer). These branches may develop into incurrent and excurrent papillae in future; the one which continues its growth may form a future chamber (CH 3).

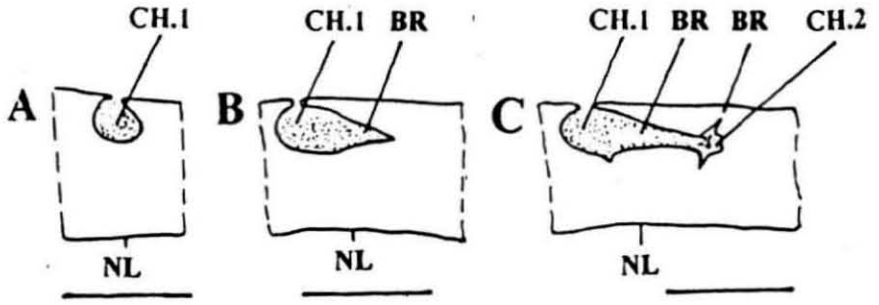
D. The two adjacent chambers, thus formed, are connected by a canal called inter chamberal canal (ICC). The interior of both chambers and canals have an etched out appearance and this is due to the chipping of calcareous matter by the sponge. The inter chamberal canal opens to adjacent chambers, through holes which are often circular. The inter chamberal canal going vertically down may appear as circular opening when viewed from above. The two new chambers, CH. 1 and CH. 2, seen in Fig. C develop further and a third chamber is formed (CH. 3). From the first chamber (CH. 1) two papillae are seen piercing the upper surface of the shell. Of these the first one is excurrent papilla (EPA) and the other the incurrent papilla (IPA). From the second chamber (CH. 2) one has pierced the upper surface and the second one is yet to pierce the upper surface. From the chambers (CH. 1 and CH. 2) branches are directed towards the inner side of the shell. These branches have not yet pierced the nacreous layer in the first chamber (CH. 1) and in the second chamber two branches are formed. Of these the first one is of papillar nature and may form either incurrent or excurrent papilla in future, while the second one is destined to become a chamber and from this chamber further growth may be effected (Scale = 1 mm throughout).

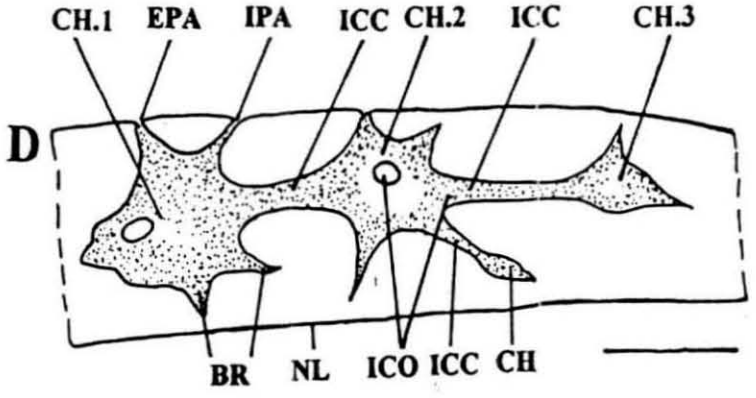
E. Longitudinal section of the shell showing three chambers, the last one (CH. 3) opens to the inner part (NL) of the shell after forming a small chamber close to the nacreous layer (NL)

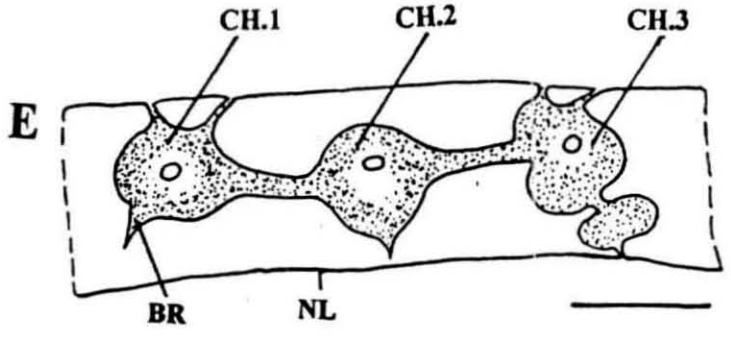
Stippled areas represent sponge growth while unstippled the original shell.

BR- branch; **CH 1-3**- chambers 1-3; **EPA**- excurrent papilla; **ICC**- inter chamberal canal; **ICO**- inter chamberal opening; **IPA**- incurrent papilla; **NL**- nacreous layer

FIGURE 5


A
CH.1
NL
B
CH.1
BR
NL
C
CH.1
BR
BR
CH.2
NL


D
CH.1
EPA
IPA
ICC
CH.2
ICC
CH.3
BR
NL
ICO
ICC
CH


E
CH.1
CH.2
CH.3
BR
NL

initial chamber thus formed (Fig. 5 B), a conical branch (BR) is formed. This branch, after some distance, enlarges to form the second chamber (Fig. 5 C, CH-2). From the second chamber a branch is formed and this, after some distance forms the third chamber (Fig. 5 D, CH-3). Thus, from the branch formed from the initial chamber, successive branches run in almost a straight-line through the interior of the shell. Canal formed in between adjacent chambers is called interchamberal canal (Fig. 5 D, ICC). This interchamberal canal open to the interior of the chamber through inter chamberal opening (Fig. 5 D, ICO), which when viewed from the interior of a chamber looks like a mere opening originating from the chamber.

As the number of chambers increase inside the shell, there is need for increased circulation of seawater for food as well as for oxygen. The water taken in has to be expelled after circulation and for this the papillae, both incurrent (IPA) and excurrent (EPA) have to be produced. Since these are to be opened out through the surface of the shell, they are formed from the upper surface of each chamber as smaller branches. These branches open out at the surface as papillae. Incurrent papillae are provided with several smaller openings (ostia) at its summit protected by brush like spicules (Fig. 6 A), while the excurrent papillae will have a single large opening (oscule) encircled by brushes of spicules at the summit (Fig. 6 B). The incurrent papillae help in the taking in of water, while the excurrent papillae in expelling water after circulating through flagellated chambers and associated canal systems (marked with arrows in figure) (Fig. 6 C). Chips removed from the interior of shell by action of sponge are expelled through the excurrent stream of water, and these chips add to the sediment load in the adjoining areas (Goreau and Hartman, 1963).

In the thinner parts of the shell (marginal areas of mussel shell) normally one tier of chambers is formed and they are confined to the middle layers of the shell. As the number or size of chambers increase, each chamber may form smaller branches from its lower part and these open to the lower side of the shell (opening into the spaces between the shell and the mantle cavity of the mussel). Since these papillae are

Fig. 6 *Cliona celata* : Diagrammatic vertical section of incurrent and excurrent papillae and the substratum of the sponge and the host

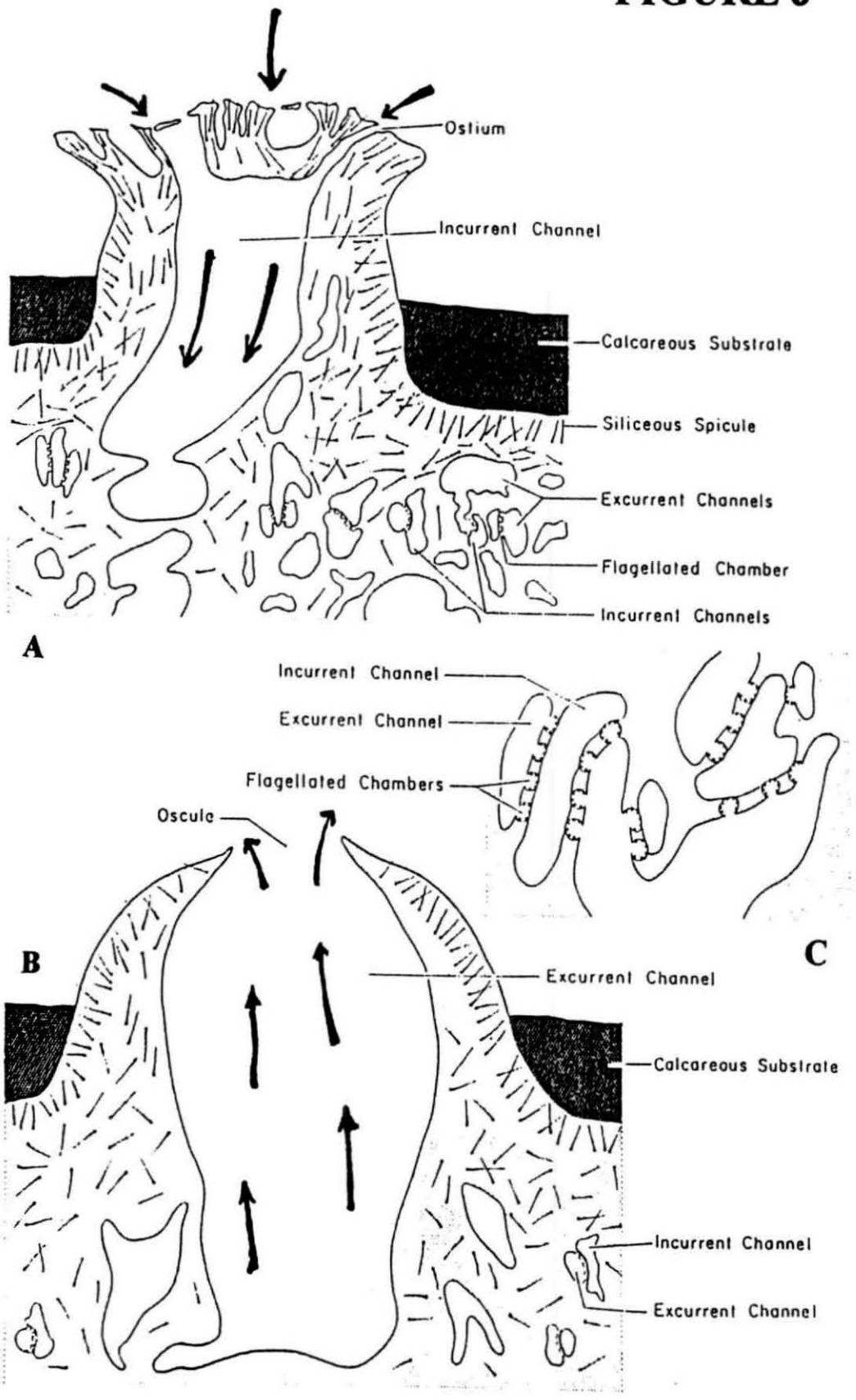
A. Section of calcareous substratum showing the longitudinal aspect of an incurrent papilla in expanded condition. The water is drawn in through the ostia situated at its mushroom-shaped summit. The water drawn in is circulated through incurrent canals, flagellate chambers, excurrent canals (channels) etc.

B. Section of the calcareous substratum showing the longitudinal aspect of an excurrent papilla fully expanded. The water already drawn in through the incurrent papilla, after circulating through flagellate chambers, is collected in excurrent canals. These excurrent canals empty the water into the main excurrent canal seen inside the excurrent papilla. The water is then ejected through the oscular opening seen at the tip of the papilla. Microchips removed by sponge from the substratum are also expelled through the excurrent stream of water.

C. Details of the incurrent canals, flagellate chambers and excurrent canals.

For A and B: Densely stippled areas represent calcareous matter; lightly stippled areas represent soft parts of the sponge and unstippled areas, the canal system. Arrows indicate the direction of water current passing through the sponge (Figure modified from Goreau and Hartman, 1963).

FIGURE 6


Ostium
Incurrent Channel
Calcareous Substrate
Siliceous Spicule
Excurrent Channels
Flagellated Chamber
Incurrent Channels
A
Incurrent Channel
Excurrent Channel
Flagellated Chambers
Oscule
B
C
Excurrent Channel
Calcareous Substrate
Incurrent Channel
Excurrent Channel

provided with brushes of spicules at their summit (Figs. 6 A, 39 A) these spicules may cause perpetual irritation to the mantle epithelium creating several pathological manifestations to the live mollusc (or any host).In the thinner areas of the shell the chambers formed are in one tier and the branch of sponge run almost in a straight line occasionally bifurcating to occupy newer areas of the shell (Fig. 21 F under *C. vastifica* and Fig. 32 C, D under *C. margaritifera*). The pores through which the incurrent and excurrent papillae project out may be in a straight line when viewed from the surface of the shell. In thinner shells like those of windowpane oyster, Thomas, (1983; Plate 1, Fig. 2) reported a linear and reticulate growth pattern for *C. vastifica* in almost all shells examined by him from Goa.

In the thicker parts (umbo proper) of the mussel shell the sponge shows a tendency to grow vertically down from the surface or from point of entry of the sponge larva. This is effected by producing chambers and canal as in the case of horizontal growth pattern described above with a difference that they are directed downwards, ie. towards the nacreous layer. Many chambers are thus formed below the initial chamber formed close to the surface. These are formed in the same pattern as seen in horizontal growth, ie. by chambers and canals. In a vertical section of the umbo proper the chambers are seen in many tiers (Fig. 2 for mussels and Fig. 9 A for *Chama* sp.). All the chambers thus formed, while growing vertically downwards, may produce lateral branches. Some such branches may accelerate horizontal growth of the sponge while others help in the production of more chambers in a three dimensional pattern. As there is an overcrowding of chambers at the umbo proper more and more incurrent and excurrent papillae may open out at the surface of the umbo region.

As the sponge grows older and voluminous, more and more space inside the shell is required and this is made possible through micro-chipping. This makes the chambers wider and wider, and finally adjacent chambers may get united either in a horizontal or in a vertical plane. Fig. 7 shows two stages in the etching away of inter chamberal septa resulting in the formation of a compound chamber (Fig. 7 A & B).

Fig. 7 Fusion of chambers to form a 'tunnel' inside the shell.

A. Fusion of chambers formed inside the molluscan shell through the etching out of inter chamberal septa (ICS). (Horizontal section through LC in Fig. 3 viewed in the direction of arrow 3). Two pairs of adjacent chambers (1 & 2 and 3 & 4) are shown. The inter chamberal septa in between chambers 1 & 2 and 3 & 4 have been etched out and thus two larger chambers are formed.

B. Two larger chambers 1 & 2, 3 & 4 have been formed by the etching out of the septa found in between. Likewise all the chambers found in the middle layers of the shell may be united into an extensive tunnel in the middle layers of the shell. Stippled areas show sponge growth and unstippled, the original shell.

CH- chamber; **ICC**- inter chamberal canal; **ICS**- inter chamberal septa

FIGURE 7

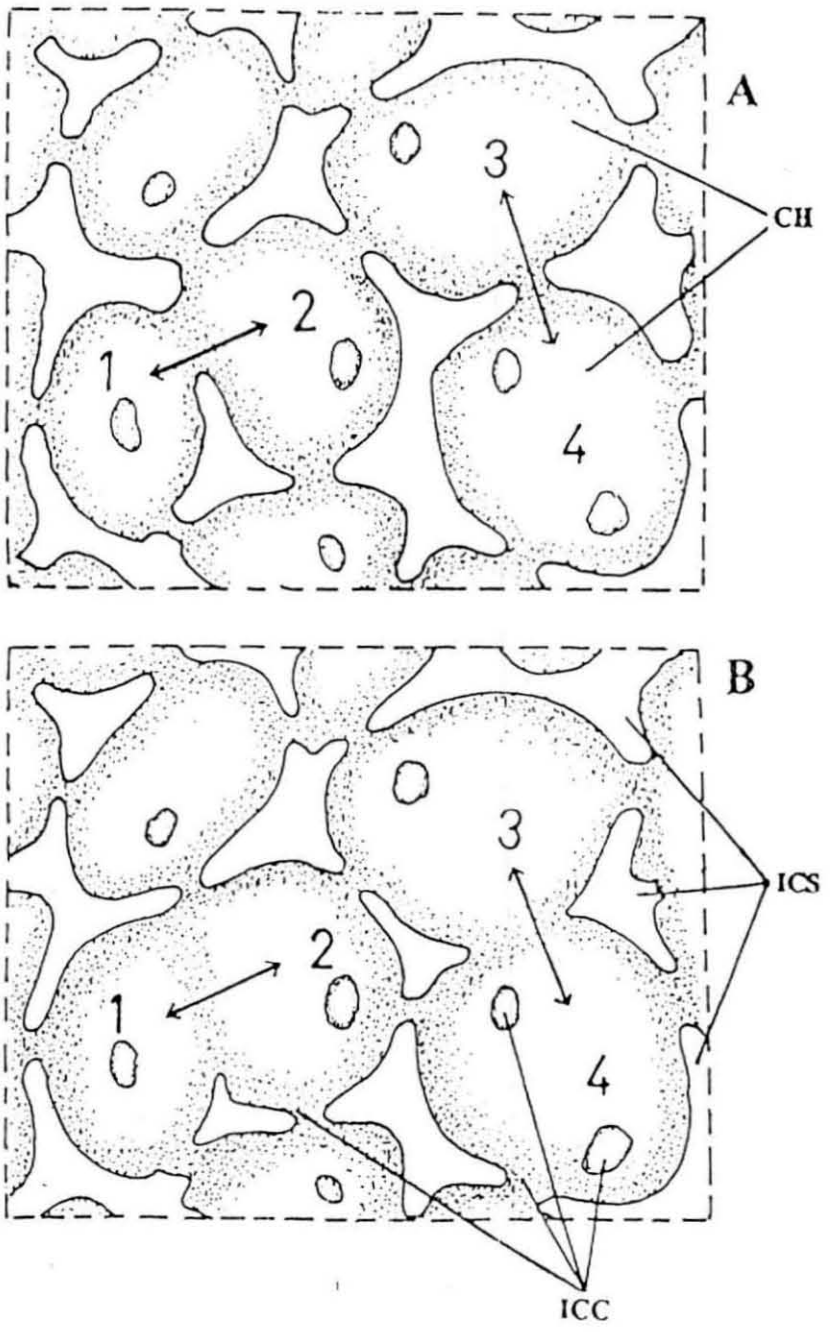


Fig. 8 Formation of a 'tunnel' inside the shell-advanced stage of boring

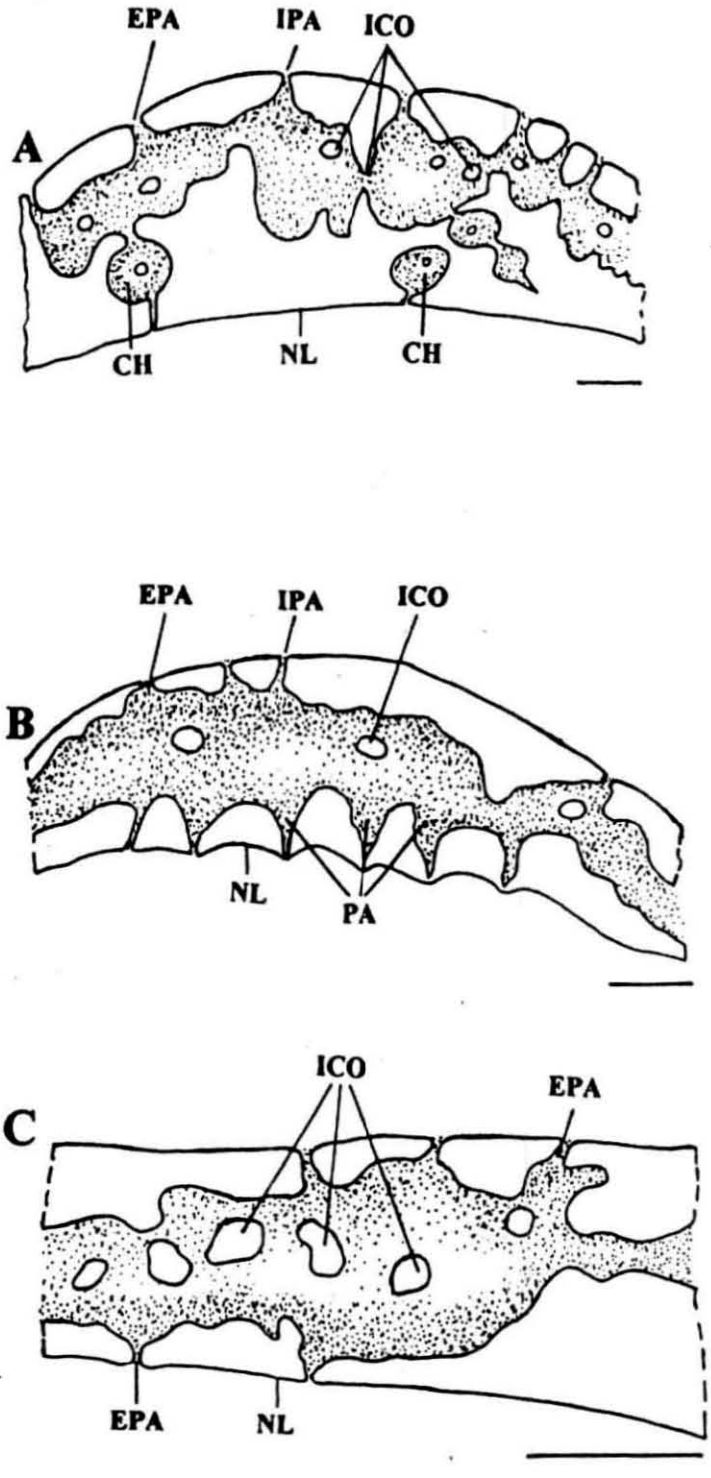
A. In thinner shells (as in mussel) the chambers formed inside are in one tier. These chambers enlarge considerably as more and more calcareous matter is etched out from the interior of the shell by the sponge. As a result both incurrent and excurrent papillae (IPA and EPA) increase in number and may be seen opening out at both surfaces of the shell. The chamber-canal arrangement, quite characteristic of the initial stages of infestation may be lost and chambers and inter chamberal canals become quite enlarged giving the appearance of a 'tunnel' (Scale = 1 mm).

B. The chamber-canal pattern of boring is completely replaced by a system of tunnels in the middle layers of the mussel shell. The papillar openings at the nacreous layer (NL) increase in number establishing contact of sponge papillae with the soft parts of the mantle. At this stage the shell may become brittle at slightest pressure (Scale = 1 mm, Shell No. 11, Kadiyapatnam).

C. Sponge infestation in an advanced stage. The tunnel made inside the shell by the sponge has enlarged considerably. The inter chamberal openings (ICO) have also become very large and irregular in out line (Scale = 1 mm; Shell No. 16, Colachel). Stippled areas show sponge growth, while unstippled, the original shell.

CH- chamber; **EPA**- excurrent papilla; **IPA**- incurrent papilla; **ICO**- inter chamberal opening; **NL**- nacreous layer; **PA**- papillar canals arising from the central 'tunnel' and piercing the nacreous layer

FIGURE 8



When boring activity advances further, a continuous tunnel may be formed inside the shell (eg. mussel, Fig. 8. A-C) leaving no signs of chambers and canals in the middle layers. The "sponge mass" inside the middle layers of the shell may communicate with the exterior through incurrent and excurrent papillae (IPA&EPA) born at the upper and lower surfaces of the shell. The diameter of the openings found at both the surfaces of the shell may increase due to the excessive etching of the canals through which the papillae protrude. But in thicker shells (eg. edible oyster *Crassostrea* spp., or sacred chank, *Xancus pyrum*) the tunnel formed by the disintegration of the middle layers may be quite wider (Fig. 9 B) occupying the entire middle layers and may be bounded by the periostracum outside and nacreous layer inside. There may be some pillar-like structures (Fig. 9 B, Plate 3. C) which are remnants of the inter chamberal septa connecting the outer and inner layers of the shell. Such shells may often crumble at the slightest pressure.

Morphology of the "sponge mass" occupying a chamber

In order to study the actual morphology of the sponge occupying a chamber (Fig. 10 A) a chamber was located and the "sponge mass" was carefully extracted (Fig. 10 C). The "sponge mass" inside the chamber is formed from a branch (BR- 1 in Fig. 10 C) running horizontally through the shell, and after forming the "mass" the branch continues its ramification (as branch 2 in Fig. 10 C). From this "mass" a branch is given off vertically downwards (Fig. 10 C, BR- 3), and another laterally (BR-4). Two papillae, one excurrent (EPA) and the other incurrent (IPA), are seen jutting out from the upper part of the chamber. These are respectively for the expulsion and intake of water. These papillae, in living conditions, project out slightly at the surface of the shell. As soon as the shell is taken out of water, these papillae may retract into the respective openings (EPA & IPA in Fig. 10 A).

The chamber was then sectioned horizontally in the direction of the interrupted line (given in Fig. 10 A) and the upper part of the chamber was then

examined in the direction of the arrow given in Fig. 10 A. The entire interior of the chamber and the canal through which the incurrent and excurrent papillae project out and also the inter chamberal canals showed a frothy appearance due to the presence of minute concavities. Each concavity represents the area from which a microchip has been removed by the sponge. The details of chips, their measurements etc. are given in the section dealing with the mechanism of boring. Etching pattern and details of branch formation from the 'mass' were studied by Thomas (1979). Four branches originating from such a 'mass' were studied and a view of the same, from above, is given in Fig. 10 E. It is seen that from the 'mass' two branches are growing vertically downwards and two laterally. All these branches, conical in shape etch out the shell as they grow. Initial branch formation by *C. carpenteri* is shown in Fig. 10 F (After Thomas, 1979). Here the branch shows the sign of expansion after reaching some distance (chamber formation ?). In both the above instances the interior of chamber enclosing the 'sponge mass' and the same enclosing the branches were equally etched out.

It is seen in the case of *Aka minuta* that the branches show the signs of etching inside but those inside the chambers may become weak in due course (Fig. 10 G, after Thomas, 1979). Schonberg (2000) similarly could not find any sharp etching inside the chambers ("erosion scars" as given in the paper) in species like *Aka mucosa* and *Zyzzya creceta* nsp., two species of boring sponges falling outside *Cliona* group. In the distal part of a new branch normally smaller chips are produced at a faster rate whereas larger chips are mined during the widening of chambers and tunnel (Rutzler & Rieger, 1973).

Stress generation to host by boring sponges

As far as the sponge ramifications are confined to the middle layers of the shell and the papillae open out only at the outer surface of the shell, the live mollusc is not much adversely affected except a weight-loss due to the removal of minute particles of shell (microchips) by the activity of the boring sponge. Since this is a sort of

Fig. 9 Comparison of boring patterns in thick shells: early and advanced stages

A. Boring pattern in a thick bivalve shell (*Chama* sp.). The sponge larva after settlement pierces and spreads downwards and sideways towards the thinner parts. As the sponge grows, further chambers and canals are formed in a three-dimensional pattern filling the entire interior. When the chambers come close to the nacreous layer and the papillae open out through pores made at the nacreous layer of the shell, they are closed by the live mantle by secreting nacreous material producing pigment plates (PPI) (Scale = 5 mm, after Thomas, 1979).

B. Sponge infestation on a chank shell (*Xancus pyrum*). Longitudinal section of the shell. Chambers increase in dimension and unite laterally by etching out of inter chamberal septa forming a continuous tunnel inside the shell. The upper and lower layers of the shell are practically spared, except for the holes made to accommodate the excurrent and incurrent papillae. The central tunnel is traversed only by pillar-like septa (PI) connecting the two outer layers of the shell. Inter chamberal openings (ICO) may or may not be seen as the calcareous material in between the chambers is completely etched out, but at some places the inter chamberal septa are retained as "pillars". Such shells may crumble at the slightest pressure (Scale = 5 mm, after Thomas, 1983). Stippled areas show sponge growth, while unstippled areas the original shell.

CH- chamber; **EPA**- excurrent papilla; **IPA**- incurrent papilla; **ICO**- inter chamberal opening; **PI**- pillar-like remnants of the shell; **PPI**- plate like pigment

FIGURE 9

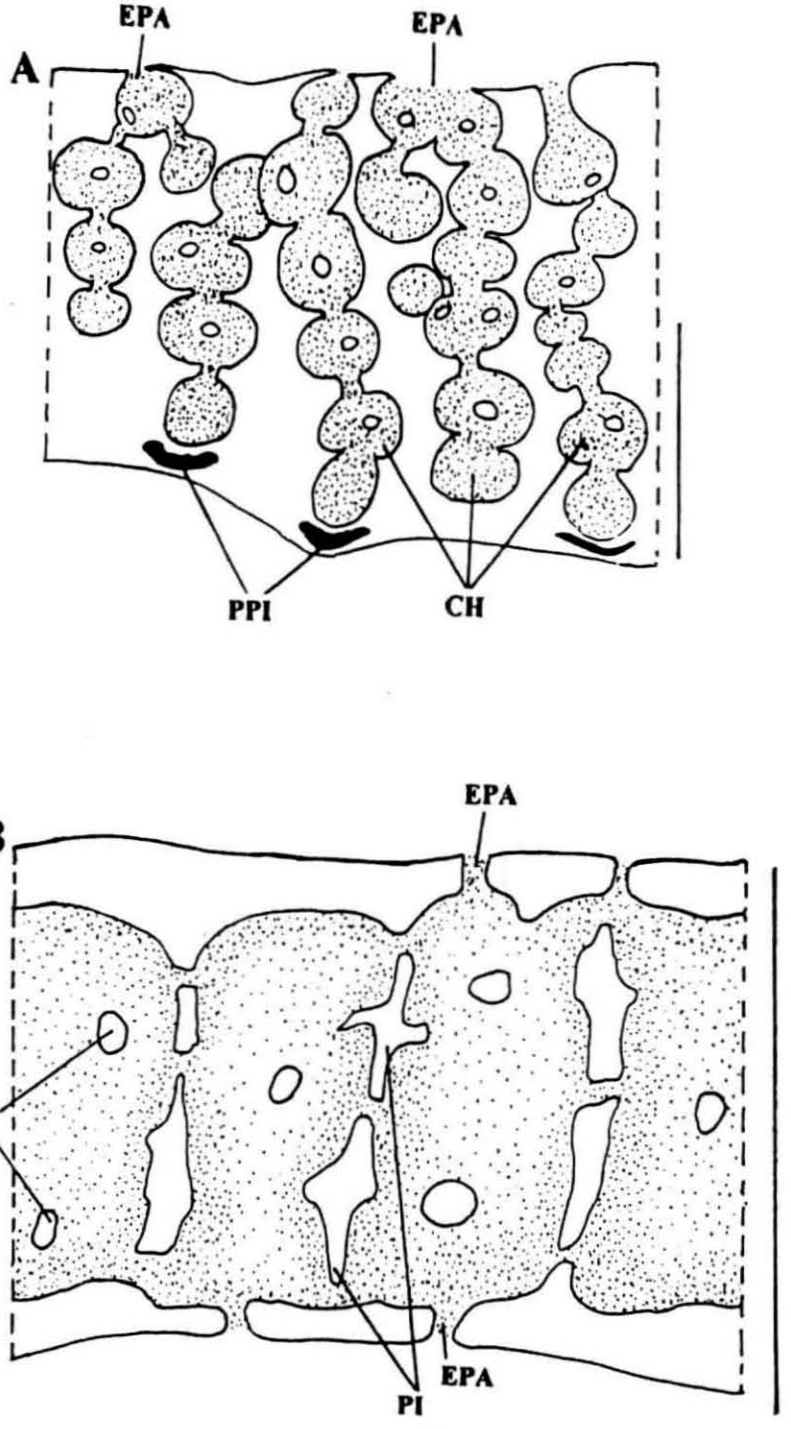


Fig. 10 Morphology of 'sponge mass' inside chambers, microchipping and proliferation inside shell

A. A chamber enlarged to show the morphology of the "sponge mass" inside a chamber. Longitudinal section of the shell showing the structure of a chamber formed inside (Scale = 1 mm).

B. Section of the chamber at a plane given in Fig A (interrupted line) and viewed in the direction of the arrow shown therein to show the etched out interior of the chamber (CH) as also of incurrent and excurrent canals (IPA and EPA) running to the surface of the shell.

C. The "sponge mass" inside the chamber (A above) is extricated to show the branches and the incurrent and excurrent papillae formed from this "mass". The 1 st branch (BR. 1), after forming the mass continues to grow as the main branch (BR. 2); the 3 rd branch (BR. 3) grows downwards and the 4 th (BR. 4) grows sideways. Incurrent papilla with several pores at the extremity (IPA) and an excurrent papilla (EPA) with a single opening at its extremity are also shown (Scale = 1 mm).

D. Pattern of etching left on the oyster shell after the settling of a clionid larva (view from above, Goreau and Hartman, 1963). Etched-out area is shown in white and the shell with stipples.

E. Spreading of boring sponge inside the substratum. A cross section of the chamber is given. Two branches are seen growing vertically downwards and two sideways. The interior of the chamber and branches have an etched-out appearance (after Thomas, 1979; Scale = 0.1 mm).

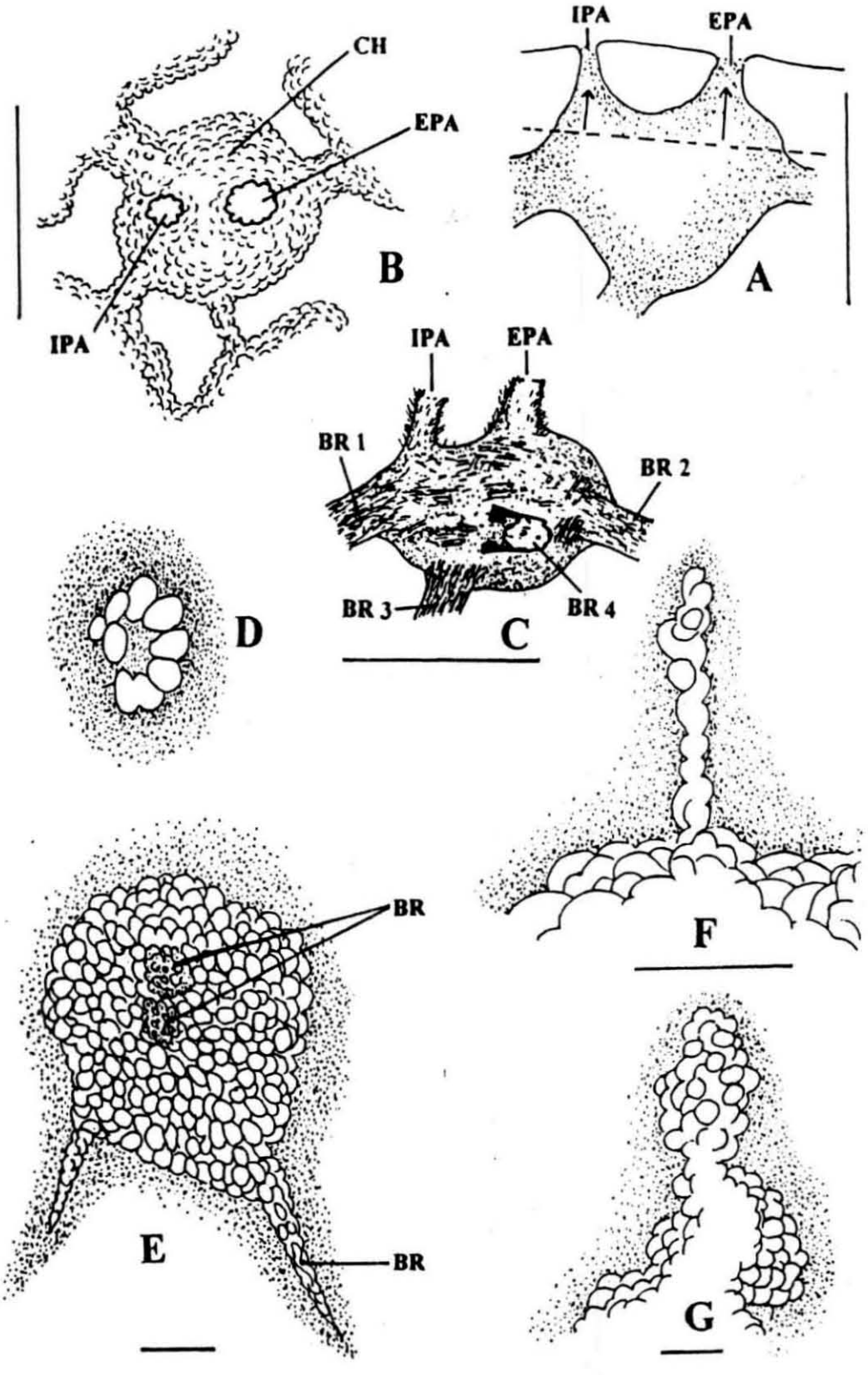
F. Branch formed from a chamber expands distally to form a new chamber. The interior is etched out completely and the branch shows a tendency to form a new chamber at its extremity (after Thomas, 1979; Scale = 0.1 mm).

G. Branch formed from a chamber (sponge: *Aka minuta* and substratum: coral). Here only the branches have an etched out interior and the chambers become internally very smooth in advanced stages (after Thomas, 1979; Scale = 0.1 mm).

(E, F, G-stippled areas represent the shell/ calcareous object, and unstippled the sponge growth)

BR. 1-4- branches 1-4; **CH**- chamber; **EPA**- excurrent papilla; **IPA**- incurrent papilla

FIGURE10



bioerosion going on at micro-level, the entire process may be termed 'bioerosion' at micro level or even "microerosion". But when the chambers and canals spread through the length and breadth of the shell, it becomes more and more brittle and with slightest pressure the entire shell would collapse.

Boring sponges may do much havoc to the live host when the sponge, in advanced stages, puts forth incurrent and excurrent papillae to the inner side of the shells. These papillae, when extended in living condition for taking in and expelling water, may even touch the soft mantle epithelium of the live mollusc. " Spicules which are arranged in a brush-like pattern at the summit of these papillae when touches the soft tissues of the mantle produce lysis of epithelium and underlying connective tissue" (Glastoff, 1964).

When the papillae pierce the nacreous layer of the shell, the live mollusc prevents it by secreting additional nacreous material and close the openings through which the papillae project out. de Laubenfels (1947) pointed out "a serious drain in oyster's energy due to the extra strain of secreting additional conchiolin to prevent the contact of the sponge with the soft tissue." As far as the mollusc is young and in its fast growing phase the openings made by sponge are repaired fast by nacreous material but when the mollusc becomes old and weak and nacreous production slows down such openings may not be repaired easily resulting in the incessant contact of sponge papillae with the soft mantle epithelium". In the actively growing phase of the mollusc, the papillae which open into the inner side of the shell through pores are prevented by a constant secretion of nacreous material by the mantle of the mollusc. Usually smaller pores through which papillae project out are easily repaired while larger ones remain open permanently. The constant secretion of shell matter around a specified point of disturbance (the opening) thus results in the formation of a blister (Fig. 11 A, B, C). When the opening made at the inner aspect of the shell of a live mollusc is repaired by nacreous material, a black patch is formed at the site of the original pore. Warburton (1958) opined that these black patches are formed by the concentration of "blood

Fig. 11 Details of blister formation in the nacreous layer of the shell of *Perna indica*

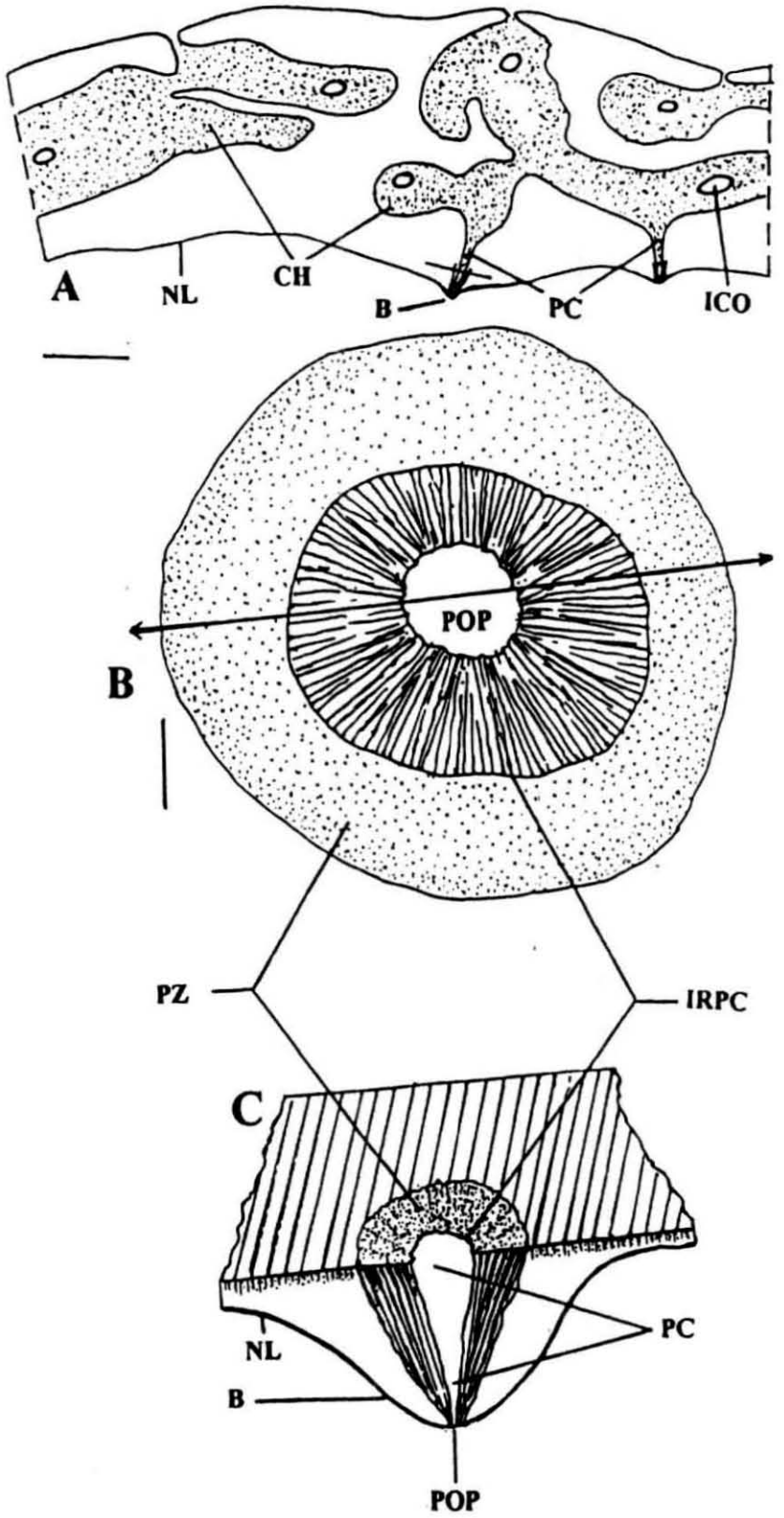
A. Longitudinal section of mussel shell bored heavily by *C. vastifica*. The papillae open to the nacreous layer (NC) of the shell establishing contact with the soft mantle epithelium. The papillar opening when closed by the secretion of nacreous material by the mussel, a blister is formed. This blister is capped with black pigment. Stippled areas show sponge growth and the unstippled, the original shell (Scale = 1 mm).

B. Area marked at the base of the blister (B), in A above, is shown in cross section. The pigment zone (PZ) and papillar canals are sloping to the terminal pore at the summit of the blister (POP). The papillar opening is now closed with nacreous material. Stippled areas show the pigment zone and the striated, its extension to the terminal pore (POP) (Scale = 0.1 mm).

C. A three dimensional figure showing the papillar canal (PC) ending at the summit of the blister (B), the pigment zone (PZ, both in cross and longitudinal sections), inner rim of papillar canal (IRPC) etc. are shown. The plain of examination is indicated by an arrow in Fig. B.

B- blister; **C**- chamber; **ICO**- interchamberal opening; **IRPC**- inner rim of papillar canal; **NL**- nacreous layer; **PC**- papillar canal; **POP**- papillar opening; **PZ**- pigment zone

FIGURE 11



cells". This pigment, in some cases (as in *C. vastifica* Fig. 11 B, C), is seen encircling the papillar canals distally (Fig. 11, B, C-PZ). Variations in the distribution of black pigment noticed are given under each species (see under "Taxonomy").

The above arrangement of blisters and pigments is generally seen in papillar canals originating singly from a chamber which is situated near the nacreous layer of the shell (as in Fig. 11 A-PC). But when the chambers remain so close to the nacreous layer as in *C. celata*, several papillar canals are produced (Fig. 12 A). A chamber with 8 papillar canals originating directly from the chamber is given in Fig. 12. B. In this case all the papillar canals may not open out through the nacreous layer but may produce only minute projections at the nacreous layer (Fig. 12 A). Those which pierced the nacreous layer and later got repaired are provided with a pigment spot at the summit (Fig. 12 C- PCP) or with a cap like pigmentation at the extremity (Fig. 12 C-PCC). In some cases the papillar canal may open outside through an opening (Fig. 12 C-PCO) without any trace of pigment.

When many such papillae with apical pigment are formed close by, it may result in the formation of a plate-like pigment zone as in the case of *C. margaritifera* (Fig. 33 D) boring into brown mussel or *C. vastifica* boring into the shell of *Chama* sp. (after Thomas, 1979).

Various physical and physiological stress situations created in living mollusc due to the infestation of boring sponges are delt in Chapter VII.

It is seen that several species of boring sponges are capable of growing independently after disintegrating the substratum totally. But all the boring sponge species infesting the brown mussel, examined during the present study, were in α stage of growth ie. sponge confined to the interior of the shell. Fusion of excurrent papillae could be noticed in one specimen of *C. celata* boring into the mussel shell from Enayam (Shell No. 38, Fig. 16 A, given under the taxonomy part of *C. celata*).

Fig. 12 "Spinous" blister formation in chambers situated close to nacreous layer: a schematic representation

A. A schematic section through lower layer of chambers. Arrows indicated the plain of observation. Papillae originating from such chambers are small, may be many in number. They produce granular to spiny prominences at the nacreous layer of the shell. LLC indicates the plain of cut made on the shell. Stippled areas indicate sponge growth and unstippled, the original shell.

B. A magnified view of the chamber in the direction of the arrow shown in Fig. A above. 8 papillar canals are seen originating from the chamber and running towards the nacreous layer and finally piercing it. These opening bear papillae (both incurrent and excurrent) inside. Each chamber is separated from the adjacent chambers by inter chamberal septa (Scale = 1 mm). Stippled areas indicate sponge growth and unstippled, the original shell.

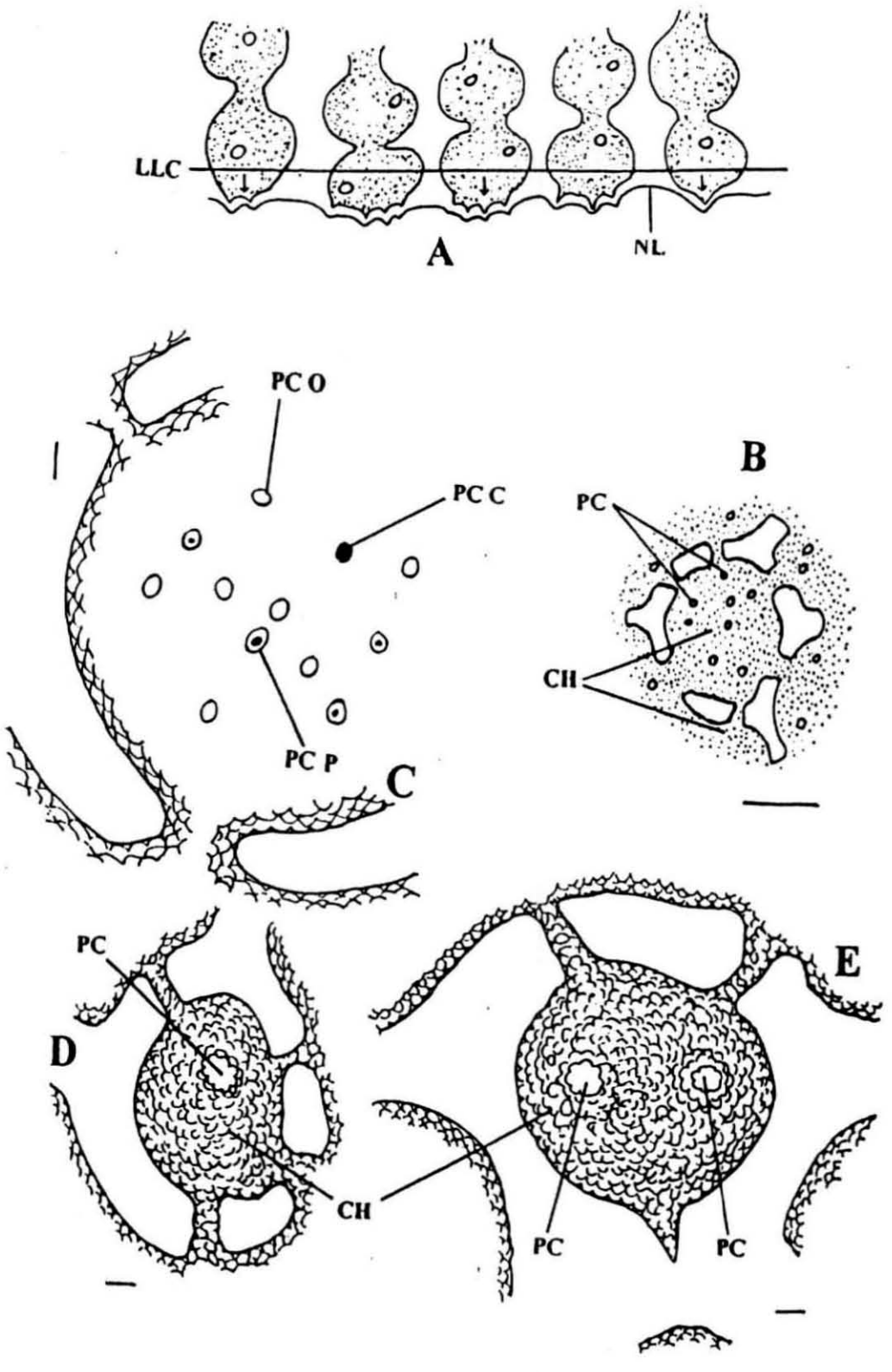
C. A chamber is more magnified to show the nature of papillar canals through which the papillae project out and touch the mantle epithelium. Some papillar canals in the figure are closed (PCC) and in some others pigment deposition is seen at its tip (PCP); some others are open (PCO) making it easy for the papillae to protrude out to the mantle cavity for taking in and expelling water. Etchings inside the chambers are partly shown (Scale = 0.1 mm, species of sponge, *Cliona celata*).

D. A chamber with one papilla opening out through the nacreous layer. The etched out interior of chamber is clearly seen (Scale = 0.1 mm, sponge *Cliona lobata*).

E. A chamber with two papillar canals opening out at the nacreous layer; the larger usually lodges excurrent and the smaller, the incurrent papillae (Scale = 0.1 mm, sponge *Cliona lobata*).

CH- chamber; **LLC**- lower layer of chambers; **NC**- nacreous layer; **PC**- papillar canal; **PCC**- papillar canal closed; **PCO**- papillar canal, terminally open; **PCP**- papillar canal, ending in pigment patch

FIGURE 12



2. MECHANISM OF BORING

1. INTRODUCTION

The unique quality of clionids is their capacity to excavate calcareous material such as coral (live or dead), shells of molluscs, calcareous worm tubes, calcareous algae, barnacle tests, limestone etc. The mechanism by which they accomplish this feat has been the subject of considerable investigations and discussions on the part of zoologists since Grant (1826) described the first clionid, *C. celata* boring in the oyster shells. Hancock (1849) believed that the excavations inside calcareous objects were the result of mechanical use of spicules. Probably the best observation of boring mechanism is that of Nassonow (1883) who experimented with *Cliona stationis* (= *C. vastifica*) boring in the oysters of the Black Sea. He collected sponge larvae and allowed them to settle on thin transparent flakes of oyster shells. Upon settling, the larvae flattened out and immediately began to bore into the calcareous matter. The first evidence of this was the appearance of a group of etched limestone lining elliptical areas on the surface of the shell (Fig. 10 D). He could notice cellular processes burrow into the shell following the course of the etched lines on the surface and eventually small chips of shell were freed and voided to the exterior of the sponge. The number of such chips freed by the end of the first day was 11-15. But the important work of Nassonow (1883) has been generally overlooked by subsequent authors (Revelle and Fairbridge, 1957; Ginsberg, 1957; Cloud, 1959) and assert that sponges bore by chemical action, and hence all excavated carbonate is removed by solution. This was quite controversial to the theory of Nassonow (1883) who advocated that the calcareous material is removed from the substratum in the form of particulate matter.

Warburton (1958) supported the views of Nassonow (1883) as he could notice the etched lines on the calcareous crystals made by *C. celata*, elliptical areas (35 - 45 microns in diameter) from which calcite fragments were removed. According to him "each fragment was bounded by several curved faces, convex or concave meeting in sharp edges." Warburton could also notice that if fragments which had been allowed to attach to cover glasses were removed gently, the cells left behind " showed a

remarkable network of thread-like interconnections and pseudopodia, often 50 μ or more long. " "These surrounded areas remain crescent in shape of the calcite particles excavated by the sponge or of the lines etched on calcite crystals". Clionids, thus, excavate their galleries in calcareous material by a relentless removal of small chips of calcium carbonate. Such chips may even be present in the oscular vicinity when the area is not much affected by strong currents.

Regarding the removal of chips, several theories have been put forward in the past. Some were of opinion that it was due to mechanical means while others held in the view that it is by chemical means. Hancock (1849) believed that the excavations inside calcareous objects were the result of mechanical use of spicules. But this theory got abandoned later as Nassonow (1883), Old (1941) and Warburton (1958) had shown that calcareous chips were removed by recently settled sponge larvae even before spicules had developed. Topsent (1888) believed that a purely mechanical action brought about by the contractability of sponge cell was sufficient to explain the destructive activities of clionids and he even ruled out the possibility of an acid in their activity. Nassonov (1924) and Vosmaer (1933) have suggested that some enzymes play a pivotal role in the attack of organic components of the molluscan shell. Moreover, the observation of Nassonow (1883) on post-larval clionids indicated that protoplasmic processes of the sponge cells are the site of boring activities of the sponge. Lettellier (1894) held the view that the contractility of cells adhering to the substratum is sufficient to dislodge particles of shell, and with the help of some threads or rods of rubber and gutta-percha cementing to the substrata and twisting in different directions, he could demonstrate the dislodging of particles similar to those produced while boring. But it is never mentioned that these particles were of identitical size or shape as compared to those produced by boring sponges.

The salient results emerged prior to 1960 on boring sponges are that Nassonow (1883), Cotte (1902) Vosmaer (1933), Old (1942) and Warburton (1958) have given considerable inputs into the mechanism of boring, and their findings could be summarized as follows:

a) Chips from calcareous substrata are removed by a localized dissolution of calcareous material at the point of contact between the cytoplasmic extension of the cell and substrate; b) no investigator has shown a lower pH at the point of contact of cytoplasmic extension of cell and the calcareous substrate; c) there is no evidence showing an increase in dissolved calcium in media in which live clionids are maintained; and d) carbonic acid produced by the sponge may be responsible for the dissolution of calcareous material leading to the formation of a chip.

Finding the limitations of light microscope Rutzler and Rieger (1973) carried out some experiments using SEM and TEM techniques to trace out the boring mechanism in *Cliona lampa* de Laubenfels in Iceland spar crystals. With a series of SEM and TEM photographs they have vividly described the various stages involved in the boring activity and this has given considerable relief to a controversy going on for the last 147 years from the discovery of the first clionid by Grant in 1826. Rutzler and Rieger (1973) could come to the following conclusions.

a. Distinct patterns of tunnels and chambers are formed inside the calcareous substrata during sponge penetration (Fig. 13, A).

b. The interior of the tunnels and chambers has a pitted appearance. Each pit represents the space from which a chip has been dislodged (Fig. 13, B, C).

c. Dimensions of the chip (average) is about 0.056 x 0.0478 x 0.032 mm (length x width x height). One side (Fig. 13, E) of the chip is convex (new cutting) while the other side (Fig. 13, F) is with several concavities (representing the etched interior of the chamber/ tunnel/ canal). The size of chips may vary considerably according to the area of their origin.

d. Apart from six cell types common to all sponges, an additional type characterised by apical filopodia is seen exclusively in boring sponges. This cell is actually responsible for the formation of the bucket-like structure (or "pseudopodial basket") (Fig. 14 PB) formed by the division

and anastomosing of filopodia and by progressive stages of plasmolysis help in etching out calcareous particles. These etching cells are of archaeocytic origin.

e. Fine cervices (approx. 0.25 µm wide) etched out by cellular activity, (Fig. 13 D, CS) result in freeing the chips from calcareous substrata (Fig. 13, D).

f. It is calculated that not more than 2-3 % of the substratum go into solution.

g. The cutting of calcareous chips is affected by chemical means, ie. by enzymatic action and their removal from the original site is affected through the contraction of filopodial basket, which is a mechanical process. These chips are then expelled out through oscules, which again is a mechanical action.

Rutzler and Rieger (1973), based on their own observations and the other details available in literature, concluded that boring sponges generally adopt the above mechanism of boring and the characteristic chips are approximately hemispherical fragments leaving a characteristic pitted appearance to the interior of the chamber / tunnel (Fig. 13, A). They stated that there is considerable size range in the expelled chips and the variation, according to them, "is most dependent on the point of origin of the chip and the micro topography of the substratum than on the species and nature of the substratum". In the distal part of a new tunnel, the cells mining small chips make the fastest progress. Larger chips are removed during widening of the tunnels and chambers. The size range of chips, pits, etc. given by Rutzler and Rieger (1973) and others may be summarized as follows for various species : (Table 1)

2. REVIEW OF LITERATURE

Several theories have been put forward in the past to explain the

Table 1. Diameter of pits and chips (in mm) produced by different species of boring sponges in various substrata (from previous works)

Pits	Chips	Species	Substratum	Source
0.045-0.070	---	*C. celata*	*Crassostrea* shell	Hartman (1958)
0.025-0.082	---	*Cliona celata*	*Mercenaria* shell	-do-
0.035-0.045	0.025-0.045	*Cliona celata*	Calcite crystal	Warburton (1958)
0.065-0.085	---	*Cliona celata*	Calcarenite	Rutzler (unpub.)
0.050-0.075	0.031-0.071	*Cliona lampa*	Shell	Rutzler (unpub.)
0.018-0.064	0.028-0.030	*Cliona lampa*	Calcite crystal	Rutzler & Rieger (1973)
0.049-0.094	---	*Cliothoosa hancocki*	Calcarenite	Rutzler (unpub.)
0.04	---	*Cliothoosa hancocki*	Calcarenite shell (unspecified)	Thomas (1979)
0.020-0.060	0.031	*Cliona orientalis*	Coral	Schonberg (2000)
0.020-0.080	0.024-0.036	*Cliona celata*	Shell	Schonberg (2000)
0.015-0.040	0.034-0.04	*Pione caesia*	Coral	Schonberg (2000)
Not recorded	0.027-0.034	*Zyzzya criceta*	Coral	Schonberg (2000)
0.030-0.070	0.04-0.05	*Aka mucosa*	Coral	Schonberg (2000)
0.020-0.090	0.013-0.064	*Thoosa hancocki*	Coral	Schonberg (2000)

Figs. 13 A-F

Microchipping of the shell: ultrastructure

A. The interior of the bored shells is enlarged to show the chambers (CH) and the canals connecting the chambers. Pits (P) are produced wherever the sponge comes into contact with the substratum. Each pit represents the area from which a chip has been removed.

B. More magnified side view of the interior of a chamber (CH) showing pits (P). A new chip (C) is formed at the bottom (of the figure) and the cervices (cs) formed by the filopodial basket are also shown on either side of the "new chip". The convex side of the new chip is not completely cut (side view).

C. More magnified view of a chamber (CH) from above showing the pits (P) formed by the sponge. Many new chips are formed; but all are retained at their places.

D. More magnified view of a "new chip" awaiting its removal from the place of origin. Cervices (cs) are seen fully encircling the "new chip". The side of this new chip facing the chamber (CH) may have concavities formed by the removal of earlier chips.

E. A "new chip' enlarged to show its newly cut convex surface (Cof). After the removal of the chip from the interior of the chamber, the "pit" or concavity, thus formed, appears concave.

F. A "new chip" enlarged to show its side facing the chamber (CH). At this side there may be many concavities and these represent the cavities or pits formed by the previously removed chips. Compare this figure with Fig. 13 D for its orientation inside the already etched out interior of the chambers.

(SEM photograph from Rutzler and Rieger, 1973)

FIGURE 13

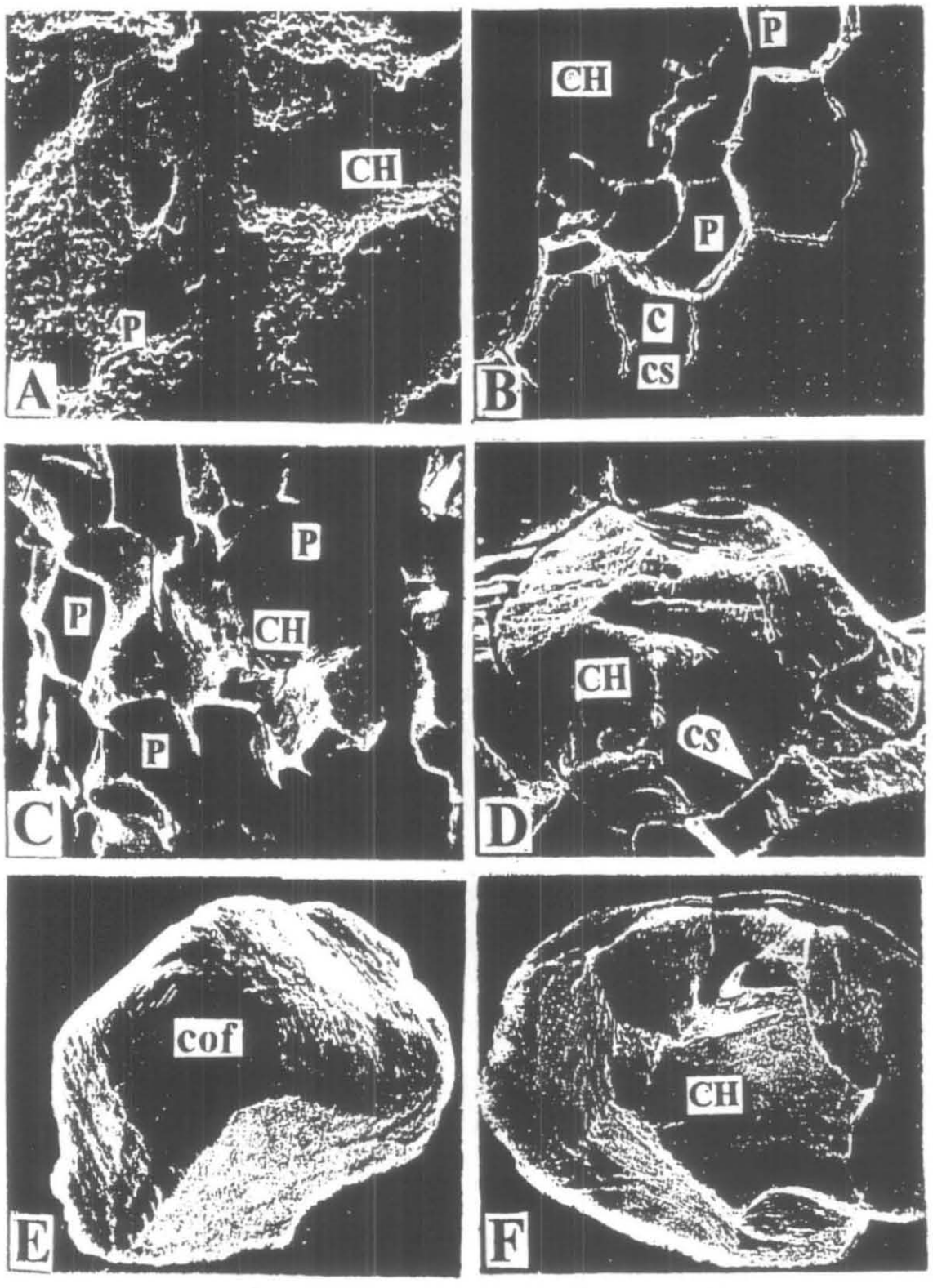
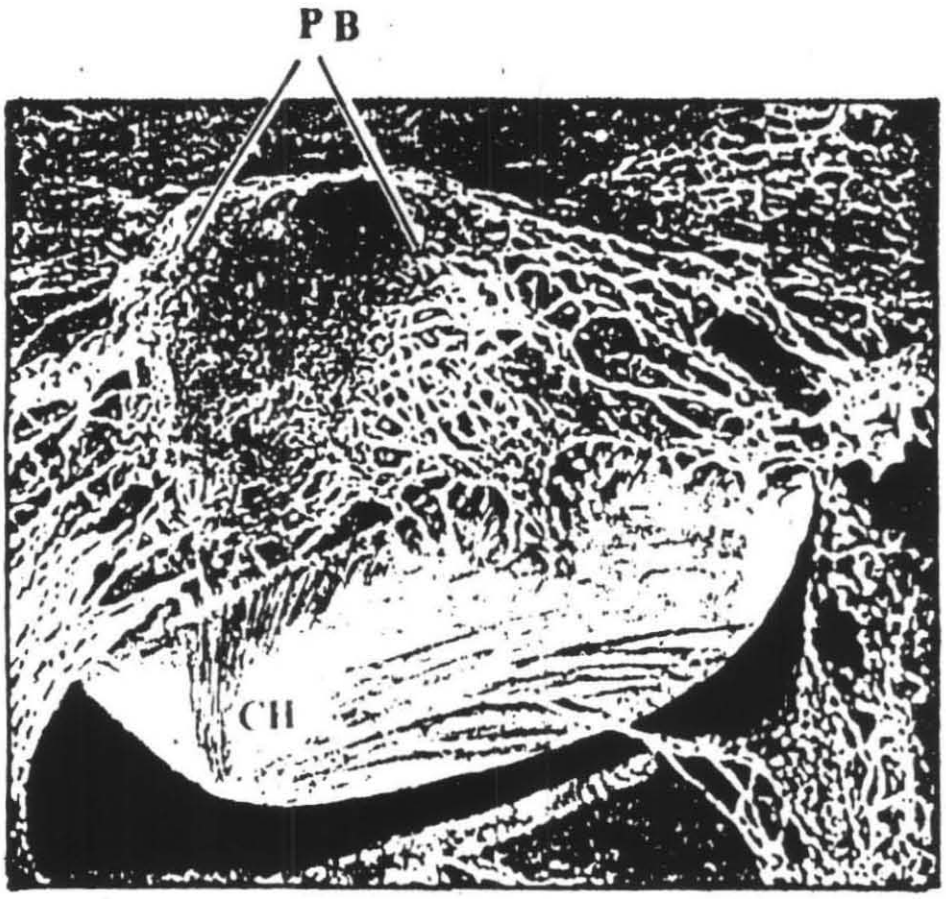


Fig. 14 Pseudopodial basket formation (After Rutzler and Rieger, 1973)

'Pseudopodial basket' (PB) covers the newly cut surface of the chip (CH) and later the chip is dislodged from its original position and sent out through excurrent canals and oscula.

FIGURE 14



phenomenon of sponge boring into calcareous substrata. The chipping of calcium carbonate particles will go on incessantly even after the death of the host (mollusc/coral). The chips could be detected in the mud fraction of sediments from the reef environment (Rutzler, 1975). Hatch (1980) demonstrated the role of carbonic anhydrase in the physiological mechanism of penetration of calcareous substrata by the boring sponge *Cliona celata.* Transmission electron microscopy of the etching areas of 11 species of clionids had shown that they have the capacity for protein synthesis, secretion, absorption and intracellular digestion (Pomponi, 1979). Pomponi also conducted cytochemical studies of acid phosphatase in the etching cells of boring sponges. Day *et al.*, (2000) measured the rate of shell erosion for two species of patellid limpets, *Patella granatina* and *P. argenvillei.*

The rate of bioerosion in one-year-old hatchery-reared shells of the black-lip pearl oyster, *Pinctada margaritifera*, by microborers and sponges was estimated (Mao Che *et al.*, 1996). Rutzler and Rieger (1973) concluded that a special type of etching cell of archeocyte origin characterised by the presence of apical filopodia is responsible for the etching process. The new chips formed inside are pulled out from the site of formation by the contraction of filopodial basket by mechanical means and are drained and expelled out through the excurrent system, which again is a mechanical process.

The nature of the substratum as well as the species of boring sponge influence the size and shape of chips. The size of microchips varies according to the location; smaller chips are released from the actively growing terminal part of the branch. They are circular to ellipsoid in outline, with concave facets on one side and a prominent convex facet on the other side. The concave facets represent parts of the pit left out by previously removed chips while the convex facets are the sites that are freshly cut by filopodial basket for making a fresh chip. Water movement, temperature, illumination, clarity of seawater, intervention by human beings and boring organisms influence the activity of boring organisms (Thomas, 1988).

The excavating activity of the sponge and the level of acid phosphatase in sponge tissues are positively correlated. This enzyme is concentrated in the cortical tissues of sponge since they are most likely to come into contact with fresh substratum. The primary mechanism of calcium carbonate dissolution involves chemical dissolution of the substrate by enzymes at the cell-substrate interface, enzymatic digestion via the lysosomal system and membranes of the etching cell processes (Pomponi, 1979).

3. MATERIAL AND METHODS

Aquarium tank of size 60 × 30 × 30 cm was used for the estimation of chip formation. A rectangular frame made of PVC tube with openings at fixed intervals was kept in the tank. Test tubes with funnels were inserted into the frame. Severely bored shells of brown mussel were kept in the funnels as shown in Plate 3. Biological filter and illumination were provided to ensure the growth of zooxanthellae which enhances the boring process. After 30 days of observation the debris collected at the bottom was examined for microchips.

4. RESULTS

The experiment on chip formation was done for a period of 30 days and the results of the experiment are shown in Table 2. The microchips expelled out through the excurrent system of sponge were collected at the bottom of test tubes were isolated and identified under a compound microscope. Measurements were taken with the help of a calibrated occular micrometer. The chips produced by *C. vastifica* and *C. margaritifera* are of uniform size (0.067 mm) while those produced by *C. lobata* measures 0.063 mm. The pits formed as a result of chip formation also varies with species. In *C. lobata* the pit size ranges from 0.029 mm to 0.076 mm and in *C. margaritifera* from 0.037 mm to 0.075 mm. The pits produced by *C. vastifica* shows a size range of 0.02 mm to 0.07 mm.

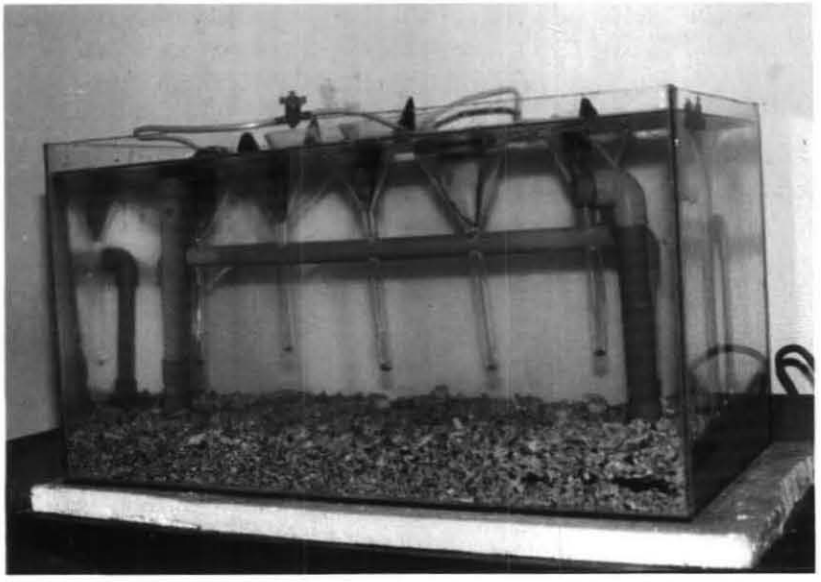


Plate 3. Experimental setup for microchip collection

Table 2. Diameter of chips (in mm) and pits made by different species of boring sponges in various substrata (present work)

Sl. No.	Pits (in mm)	Chips (in mm)	Species	Host
1	0.02-0.07	0.067	*Cliona vastifica*	Brown mussel
2	0.029-0.076	0.063	*Cliona lobata*	Brown mussel
3	0.037-0.075	0.067	*Cliona margaritifera*	Brown mussel
4	0.025-0.072	No data	*Cliona carpenteri*	Brown mussel
5	0.02-0.075	No data	*Cliona celata*	Brown mussel
6	0.025-0.04	No data	*Thoosa armata*	Brown mussel
7	0.037-0.063	No data	*Thoosa hancocki*	Rock oyster
8	0.03-0.05	No data	*Aka minuta*	Brown mussel
9	0.016-0.024	No data	*Halina extensa*	Rock oyster

5. DISCUSSION

The average size of chips generated by *C. lampa* in Iceland spar was 56×47×32 µm (Rutzler and Rieger, 1973). The chipping rate of clionids can be calculated indirectly from the calcium content of the seawater and also from the weight loss of the substratum at periodic intervals. Rutzler and Reiger could experimentally ascertain that about 2-3 % of the substratum was removed in the form of solution and 1 mg dried tissue of *C. lampa* was capable of dislodging 16 mg calcium carbonate particle over a period of one year. Chemical etching detaches a hemispherical multifaceted chip of calcium carbonate of about 0.04-0.06 mm size which is then mechanically removed through the excurrent canal system (Pomponi, 1979). The rate of bioerosion in one-year-old hatchery shells of *Pinctada margaritifera* was 36 times higher than in natural populations (Mao Che *et al.*, 1996).

When boring into the nacreous region they penetrate deep into the shells leaving etch marks in the form of striations following the arrangement of the aragonite platelets. Sponges can be well defined as successful borers, and exploit all the available substrate. The highest rate of erosion of *Patella granatina* shells (36 %) was recorded when the shells were grazed by a co-occurring limpet, *P. granularis*, but in their absence erosion was only 15.2 %. This shows that coexistence with other species enhances the bioerosion rates in some molluscs (Day *et al.*, 2000). Hatch (1980) demonstrated that the concentration of carbonic anhydrase is related to the excavating activity of the sponge *C. celata*.

Bioerosion rates determined from short duration experiments (Neumann, 1966) were found to be erroneous by Rutzler (1975) who showed that sponge bioerosion is rapid for the first three to six months, while the sponge is becoming established within the substrate, but subsequent bioerosion proceeds much more slowly. In the present study the chip size of *C. lobata*, C. *vastifica* and *C. margaritifera* were of almost uniform size.

3. TAXONOMY OF MUSSEL BORING SPONGES

1. INTRODUCTION

In order to make a comprehensive list of boring sponge species occurring in the study area, shells of brown mussels as also of other molluscs were examined and a total of 10 species were collected. Description is provided for all species in the following account.

2. REVIEW OF LITERATURE

Thangavelu and Sanjeeva Raj (1988) described the boring and fouling organisms of the edible oyster *Crassostrea madrasensis*. Bower *et al.*, (1994) studied the shell burrowing sponges of mussels from North America. Predation by various invertebrates on the burrowing sponge, *Cliona celata*, was investigated both in the laboratory and field (Guida, 1976) in order to determine the importance of predation to sponge population and the adaptive advantage of burrowing to escape predators. Alagarswamy and Chellam (1976) conducted experiments to study the fouling and boring organisms and mortality to pearl oysters at Veppalodai farm in the Gulf of Mannar. Rutzler and Bromley (1981) identified a new species (*Cliona rhodensis*) of boring sponge from the Mediterranean Sea.

Bottjer (1981) discussed the role of periostracum of the gastropod *Fusitriton oregonensis* as natural inhibitor of boring and encrusting organisms. The incidence of *Cliona celata* boring into the shell of *Patella vulgata* in Orkney was studied by Baxter (1984). The boring organisms in living gastropods at fifteen sites around Guam were studied by Smyth (1990). Wesche *et al.*, (1997) discovered two species of clionid sponges from the shells of living and dead Sydney rock oyster, *Saccostrea commercialis*. Schleyer (1991) worked on the shell borers on the oyster, *Striostrea margaritacea* in Natal.

The periostracum of the gastropod *Hemifuscus pugillinus* acts as the

natural inhibitor of boring and encrusting organisms (Anandakumar and Ayyakkannu, 1991). Rosell and Uriz (1992) made studies to find out whether zooxanthallae and the nature of the substratum affect survival, attachment and growth of *Cliona viridis.* Two species of boring sponges belonging to the order Haplosclerida, viz. *Siphonodictyon mucosum* Bergquist and *Siphonodictyon coralliophagum* forma *tubulosa* were reported from Maldives (Thomas, Bakus and Wright, M.S) and thus two haplosclerid sponges, as boring forms, are added to the Indian Ocean list of boring sponge species. The ecological and morphological relationships of two clionid sponges, *C. vastifica* and *C. lobata* collected off the coast of Blanes were compared by Rosell (1994). Mao Che *et al.*, (1996) studied the composition, distribution and infestation sequence of organisms that destroy the commercially valuable shells of the black-lip pearl oyster *Pinctada margaritifera.* A comparative study of the process of molluscan shell erosion in two patellid limpets was made by Day *et al.*, (2000). Clavier (1992) reported an infestation of *Haliotis tuberculata* shells by *Cliona celata* and *Polydora* sp. in their natural environment.

3. MATERIAL AND METHODS

The details regarding collection methods, laboratory examination, spicule preparation, examination of sections, literature referred to etc. are given in depth in the part dealing with "Material and methods" as given in Chapter 1.

Alectona millari Carter (1879), a sponge common to the corals of Atlantic and Mediterranean Sea, is recorded here from the Indian Ocean, boring into brown mussel. *Thoosa hancocki* Topsent is recorded here from brown mussel shell and was earlier known only from shells other than those of brown mussel. *Halina extensa* is here confirmed as a boring sponge and the same is established with salient illustrations. Analyses of the shells revealed that the above said nine species, seen in brown mussel, are quite common in other shells also, but one species of the order Carnosida, viz. *Halina extensa* (Dendy), was found distributed only in the rock oyster shell at Vizhinjam (Station 1).

4. RESULTS

3. 1 LIST OF SPECIES IDENTIFIED FORM THE SOUTHWEST COAST OF INDIA AND A KEY FOR THEIR IDENTIFICATION ARE GIVEN BELOW

Source: Brown mussel

Phylum Porifera Grant

Class Demospongiae Sollas

Order Hadromerida Topsent

Family Clionidae Gray

Cliona group:

Genus Aka de Laubenfels

1. *Aka minuta* Thomas

Genus Cliona Grant

2. *Cliona celata* Grant
3. *Cliona vastifica* Hancock
4. *Cliona lobata* Hancock
5. C*liona carpenteri* Hancock
6. *Cliona margaritifera* Dendy

Thoosa group

Genus Alectona Carter

7. *Alectona milliari* Carter

Genus Thoosa Hancock

8. *Thoosa hancocki* Topsent
9. *Thoosa armata* Topsent

Source: Other molluscs

Order Carnosida Carter

Family Halinidae de Laubenfels

Subfamily Halininae de Laubenfels

Genus Halina Bowerbank

10. *Halina extensa* (Dendy)

3. 2. Key to the identification of various species

1. Megascleres diactinal 10

2. Megascleres monactinal (tylostyle) 4

3. Megascleres tetractinal (triaenes) 11

4. and with spirasters or with bacilliform microscleres 5

4 A. and with astrose microscleres ... 9

5. with spirasters as microscleres 6

5 A. with bacilliform microscleres and acanthoxeas - *Cliona carpenteri*

6. with or without spirasters and with hair-like oxeas - *Cliona celata*

6 A. spirasters distributed rather abundantly 7

7. with spirasters and acanthoxeas 8

7 A. with long spirasters but without acanthoxeas - *Cliona lobata*

8. with acanthoxeas gradually grading into spirasters - *Cliona margaritifera*

8 A. with acanthoxeas and smaller spirasters - *Cliona vastifica*

9. amphiasters, oxyasters and oxeas for microscleres - *Thoosa armata*

9 A. amphiasters only for microscleres - *Thoosa hancocki*

10. Spicules smooth diactinal forms (oxeas), no microscleres - *Aka minuta*

10 A. Spicules tuberculate diactenes and with microscleres - *Alectona millari*

11. Spicules triaenes with dicho- modifications and with microscleres- *Halina extensa**

**H. extensa*, though not found infesting the mussel shells, is included here for the comprehensiveness of the key.

Family Clionidae (Gray, 1867)

Diagnosis: Excavating Hadromerida with α β and γ growth forms (ie. cryptic papillate, cryptic encrusting due to fusion of papillae externally and free living after disintegrating the substratum totally). Spicules: tylostyles, styles and/ or oxeas and spirasters, amphiasters and/ or microrhabds.

de Laubenfels (1936) divided the family into two sub-divisions (sub-families ?). The first division is exemplified by the typical genus *Cliona* Grant and the second by *Thoosa* Hancock. Old (1941) recognised these two groups as A and B respectively. In *Cliona* group the microscleres are simple spiraster type while in *Thoosa* group they are elaborate astrose type; only α stage is noticed in this group. Two genera of *Cliona* group are represented here. They are *Aka* de Laubenfels with one species and *Cliona* Grant, with five species. Of *Thoosa* group, two genera are represented, viz. *Thoosa* Hancock *and Alectona* Carter (with two and one species respectively).

3. 3. Description of species

1. *Aka minuta* Thomas
(Figs. 10 G, 15)

Aka minuta Thomas, 1972, p. 343, pl. 2, figs., 4, 4 A; Thomas, 1973, p. 59, pl. 3, fig 9; Thomas, 1979, p. 34, pl. 2, fig. 13; Thomas, 1979 A, p. 63, pl. 3, fig.18; Thomas, 1979 B, p. 170, fig. 1C; figs. 3 A, D; pl. 3; Thomas, 1989, p. 160; Thomas *et al.*, 1993, pp. 144-156

Material: Examined five brown mussel shells from Station II, (Map I). It is here recorded as a pest of brown mussel for the first time.

Depth: Mussel beds, 5-8 meters.

Colour: Not recorded in living condition, light brown when dry.

Description: Surface of shell perforated by openings varying in diameter from 0.1 to 0.6 mm (Fig. 15 A). The incurrent (IPA) and excurrent (EPA) papillae project out through

these openings. In living condition (Fig.15 A) they are not contractile. These papillae may be up to 4 mm and have a papery consistency. Excurrent papillae have a single opening at the tip while the incurrent papillae have several openings.

Vertical section of the shell shows that the chambers (CH) formed by this species are in one tier as seen in *Cliona* spp. Chambers usually occupy the middle layers of the shell almost to about 2/3 its total thickness. Excurrent papillae are usually seen opening to the outer surface of the shell (Fig. 15 C).

A horizontal section of the shell taken through LC (as given in Fig. 3 in the direction of arrow 3) shows that the chambers (CH) formed are irregular in outline and are without any distinct interchamberal septa. The interchamberal canals (ICC) are also irregular as compared to those seen in *Cliona* spp. (Fig. 15 B). The etchings are weak (Fig. 15 D) inside the chambers (CH) and also inside the interchamberal canals (ICC).

"Sponge mass" growing inside a chamber was then extracted to study its morphology (Fig. 15 E). It could be seen that the "mass" inside the chamber originates from a branch as in *Cliona* spp., and three branches (BR. 1-3) are formed from this 'mass'. There is every possibility that BR. 1 may grow further and form another chamber, while BR. 2 and BR. 3 are destined to produce lateral chambers. Oxeas are seen projecting out of this mass and also from all branches formed.

Spicules: Oxeas 1. (Fig. 15 F1): Sharply and gradually pointed, well developed forms with an angle at the centre; size, 0.06- 0.134 mm x0.002-0.006 mm (length x width).

Remarks: The nearest relative of this species is *Aka nodosa* (Hancock, 1849) boring into the shells of the giant clam *Tridacna gigas*. Thomas (1972) recorded larger chambers (5 x 4 mm) made by this species in corals from the Gulf of Mannar. Since then this species was recorded in corals from different parts of the Indian Ocean (Seychelles, Inhaca, Mamboneis and Lakshadweep). It is the first record of this species as boring sponge of a mollusc (*Thais rudolphi*) by Thomas *et al.*, (1983), and it is here recorded as a pest of brown mussel.

Thomas (1979 B, Fig. 10 D is reproduced here in Fig. 10 G) could record the etchings only at the site of newly formed branches while the interior of the chambers is with weak etchings. *Aka mucosa* recorded by Schonberg (2000) from Great Barrier Reef of Australia also showed less pronounced pits inside chambers, and attributed it to be a generic character of the genus *Aka*. Chips produced by *Aka mucosa* measured 41-49 μm (for two specimens from Great Barrier Reef, Australia).

Distribution: Widely distributed in the Indian Ocean.

Fig. 15

Aka minuta Thomas

A. Surface of the mussel shell (Shell No. 10, Enayam) showing the opening through which the excurrent and incurrent papillae project out at the surface (see Fig. 3, viewed in the direction of arrow marked 1) (Scale = 1 mm).

B. Section through the layer of chambers (LC in Fig. 3 viewed in the direction of arrow 3). Chambers formed are quite irregular but some are oval in outline (Scale = 1 mm). Stippled areas represent sponge growth while unstippled, the original shell.

C. Vertical section of shells showing the arrangement of chamber inside. Only one tier of chambers is seen (Scale = 1mm). Stippled areas represent sponge growth while unstippled, the original shell.

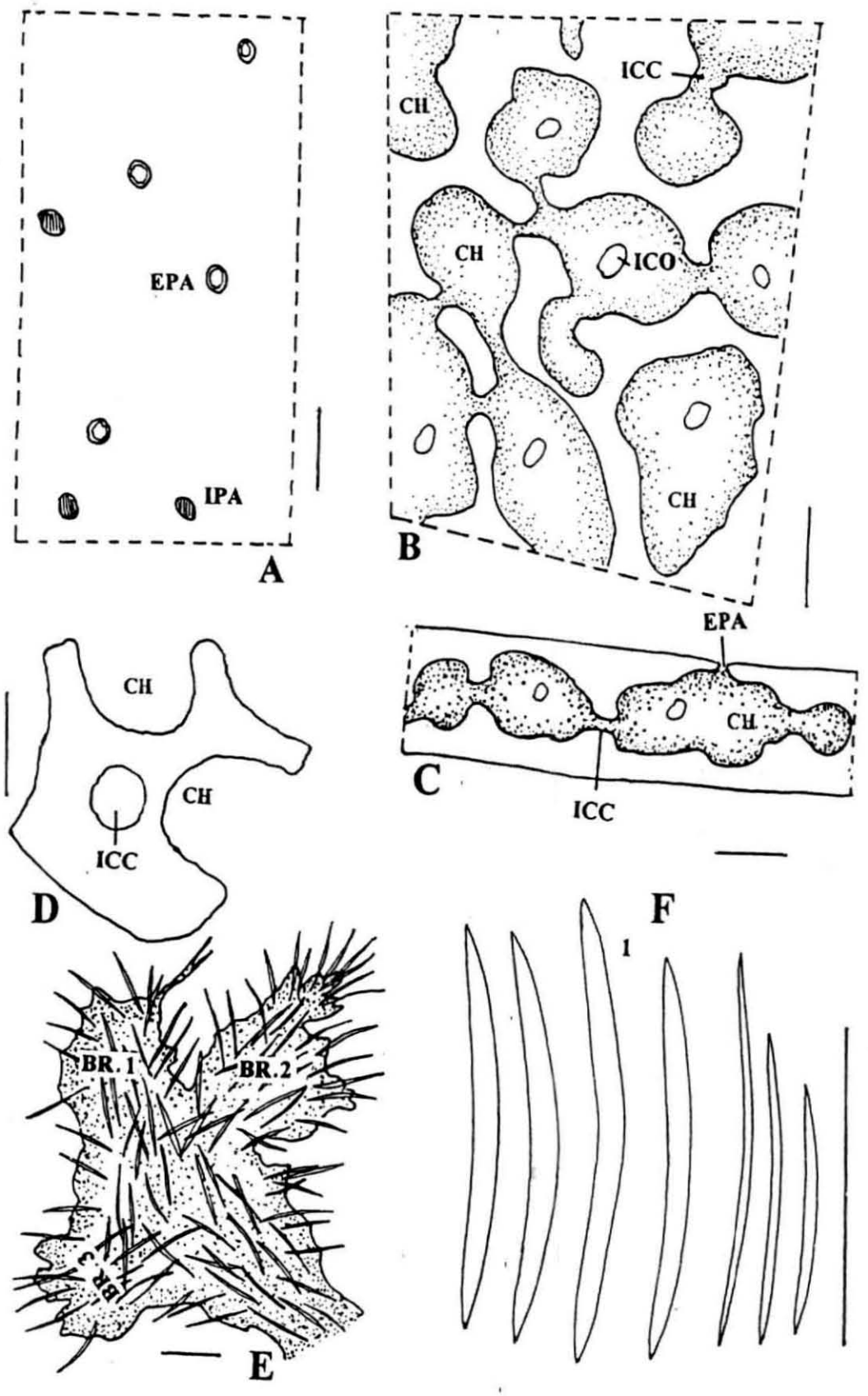
D. Horizontal section of a chamber (CH) and inter chamberal canal (ICC) showing the weak etching in the interior (Scale = 1 mm).

E. Sponge mass extricated from a chamber showing three branches (Branch 1, 2 & 3) formed from the mass inside (Scale = 0.1 mm).

F. Spicules: 1. Oxeas: thin, thick and centrangulated (Scale = 0.1 mm).

BR 1, 2, 3- branches 1-3; **CH**- chamber; **EPA**- excurrent papilla; **ICC**- interchamberal canal; **ICO**- interchamberal opening; **IPA**- incurrent papilla

FIGURE 15



2 . *Cliona celata* Grant
(Fig. 16-20, Pl. 4 A-C)

Restricted synonymy:

Cliona celata Grant, 1826, p. 79 & figs; Topsent, 1900, p. 32, pl. 1, figs. 5, 6- 9, pl. 2, fig.1 (synonymy); Vosmaer, 1933, pp. 349-383 (synonymy); Old, 1941, p. 8, pls.1, 2, 3, 7& 8; Hartman, 1958, p.16; Bergquist, 1961, p. 44; Little, 1963, p. 57; Hartman, 1964, p. 2, pl.1; Thomas, 1972, p. 344, pl. 1. figs. 5, 5 A, 5 B, 5 C; Thomas, 1973, p. 60, pl. 3, fig.10 (synonymy); Rutzler, 1973, p. 624, fig. 1; Thomas, 1979, p. 35, pl. 2, fig.15; Thomas, 1979 B, p.171, fig.1 D, fig. 3. G, H-L, M, pl. 4, figs. 4-8, pl. 5, figs.2, 4-6, 9, pl. 6. figs.1, 3-5, pl. 7, figs. 3-6; Hoshino, 1981, p. 233, fig. 24; Thomas, 1986, p. 317; pl. 6, fig. 12; Thomas, 1989, pp. 150-161; Thomas *et al.*, 1983, pp.1-13, Thomas, 1990, pp. 202-216; Thomas *et al.*, 1993, pp. 145-156; Schonberg, 2000, p. 174, fig. 3 C, 6, pl. 2 fig. 13, pl. 6, figs. 31-33; pl. 10, fig. 57.

Material: Examined 41 of bored mussel shells from different stations (Station I-VI, Map 1). Apart from shells of mussel and other bivalve shells gastropod shells collected were also examined.

Depth: Mussel beds distributed in 5-8 metres.

Colour: Green to golden yellow or red in living condition.

Description: The surface of the shell is perforated by circular to oval openings with a diameter varying from 0.037 to 1 mm. Smaller openings lodge incurrent papillae (diameter, 0.037 mm to 0.28 mm), while the larger openings, the excurrent papillae (diameter 0.037 mm to 1 mm) (IPA and EPA in Fig. 16 A). In living condition these papillae project out to about 4 mm from the surface of the shell and retract into the shell when taken out of water. Incurrent pores, in groups, are located at the summit of incurrent papillae, and excurrent openings (oscules) are solitary and terminal, one per excurrent papilla. These oscules may contract along with papillae and occupy a position flush with the surface when taken out of water. The chambers formed inside the shell are in many tiers at the umbo part, while in the thinner parts of the shell they are in one tier.

Skeletal arrangement inside the chamber is irregular, whereas in papillae it is quite dense towards the base and less dense and radial at the tips. The chambers

(CH), in a horizontal section of the shell through ULC (Fig. 2, viewed in the direction of arrow 3) appears almost pentagonal to hexagonal in outline with the diameter varying from 1 to 2 mm (Fig. 16 B). In gastropod shells the chambers may be larger, and a cross section of the spine of *Murex* sp. is given in Fig. 16 C, where the larger diameter of chambers comes to about 6 mm. These chambers are interconnected with adjacent chambers through interchamberal canals (ICC) and the original shell (unbored part) is retained in between the adjacent chambers as inter chamberal septa (Fig. 16 D, ICS). This sort of a chamber -interchamberal canal pattern may get exaggerated much as the borings become extensive. These chambers, interchamberal canals and the interchamberal septa may disappear gradually and a continuous tunnel-like cavity may be formed inside the shell in advanced stages of boring (Fig. 17 A, B, C).

The interior of the chambers (CH), the interchamberal canals (ICC) and the papillar canals, when examined under high power of microscope, present an etched out appearance (Fig. 16 D). Even the minute branch formed by the sponge from a chamber may have such etchings inside (Fig. 16 D, BR). Each such etched out pit may represent the area from which a microchip has been removed by the activity of the sponge. The diameter of the pits may vary from 0.02 mm to 0.075 mm.

In advanced stages of infestation chambers may be formed very close to the nacreous layer of the shell (Fig. 18 C), or they may be confined to the middle layers (Fig. 17 A-C) only. At this stage the sponge may put forth papillae through the inner side (nacreous layer of the shell) by piercing the nacreous layer. In this process two types of papillar canals may be formed.

1. very strong papillar canals, one per chamber (Fig. 17 A, B, C), and
2. minute papillar canals, many in number, originating from a chamber situated very close to the nacreous layer (Fig. 18 C). In the former case the papillae may pierce the nacreous layer and open out for the intake and expulsion of water. These papillae, when expanded, may even touch the soft parts of the mantle epithelium creating perpetual irritation to the live mollusc. The mollusc, in order to prevent it,

may secrete additional nacreous material and 'repair' the holes made by sponge in the inner aspect of the shell. Due to repeated repair a blister may be formed at the inner side of the shell at the point of repair. As far as the animal is healthy and young, the holes made by sponge are repaired but when it becomes weak and old the openings made may remain as such forever. When the opening is repaired, a black 'pigment' is deposited at the site of the original pore (Fig. 17 C, a closed opening with 'pigment' and four active papillar openings are shown at the nacreous layer of the shell).

3. In the second category mentioned above, several shorter papillae are formed from a chamber (Fig. 18 C, viewed in the line of arrow directed downwards from LLC, diagrammatic representation). Magnified view of the interior of a chamber is given in Fig. 18 A, which shows that as many as 16 short papillae are produced from a chamber and its magnified view is given in Fig. 18 B.

These papillae, being very small, the openings made at the nacreous layer are also very small and hence by the 'repair' of these minute pores with nacreous material, only small, spinuous projections are formed, in other words, these are minute blisters with sharply pointed tips (spine). Here also, as in the normal blister, pigment formation may be seen. In advanced stages, the pores through which these minute papillae project out may also be seen at the tip of these spinuous structures. When pigments from adjacent papillae merge together, a "plate-like pigment" may even be formed (PPI in Fig. 33 D under *C. margaritifera*). View of such spine-like structure as seen from the nacreous layer (arrow marked upward in Fig. 18 C) is given in Fig. 18 D. Such spinuous structures, when concentrated at the nacreous layer, may give a characteristic roughness to the nacreous lining inside the shell.

Pigments seen at the summit of larger blisters are of many types, circular, stellate etc. (Fig. 19, 1 to 4).

General morphology, anatomy, physiology and development of this species have been extensively studied (Topsent, 1900; Hartman, 1958; Goreau and

Hartman, 1963). Details on growth patterns, "sponge mass" inside the chambers, branch formation and their spreading inside the shell are given in the chapter dealing with "pattern of boring".

Spicules: 1.Tylostyles, (Fig. 20, 1): Smooth and slightly curved at the first half, tips sharply pointed, head trilobed to oblong, size; 0.08 -0.29 mm x 0.002 -0.013 mm (length x width).

2. Oxeas (Fig. 20, 2): Very rare and with hair-like dimensions. This spicule was present only in two specimens examined, size, 0.08 -0.14 mm.

Remarks: Only α stage is seen in the Indian seas. In a few mussel shells examined, some papillae showed the signs of fusion at the surface of the shell (Fig. 16 A). No spiraster could be seen in the present specimens though the presence of spirasters was reported by many in the past.

Distribution: It is a species with cosmopolitan distribution.

Fig. 16

Cliona celata Grant

A. Upper surface (US in Fig. 2) of mussel shell showing the distribution of incurrent and excurrent opening through which the papillae (IPA and EPA) project out; excurrent papillae with single opening at the summit. Some excurrent papillae are fused with the adjacent ones (β stage) (Shell No. 38; Enayam) (Scale = 1 mm).

B. Horizontal section of the shell at ULC (See Fig. 2, viewed downwards in the direction of arrow 3) to show the structure of chambers (CH), inter chamberal septa (ICS), inter chamberal canals (ICC) etc. Circular openings (1 or 2) seen inside the chambers are the openings of the interchamberal canals (ICC) (Shell No. 27, Enayam) (Scale = 1 mm). Stippled areas represent sponge growth while unstippled, the original shell.

C. Cross section of the spine of *Murex vergenius* (Vizhinjam, No. 3) showing large chambers made inside by *Cliona celata*. Stippled areas show sponge growth while unstippled, the original shell (Scale = 2 mm).

D. Magnified view of a chamber. The interior of the chamber gives a frothy appearance due to the removal of microchips. Each concavity represents the place from which a microchip is removed. In between adjacent chambers the original shell is retained and is marked as interchamberal septa (ICS). These septa may get pierced by the formation of a new branch (BR) from the "sponge mass" inside the chamber. When interchamberal septa is thick these opening form a canal (interchamberal canal, ICC) with opening at either end called the interchamberal opening (ICO) (Scale = 0.1 mm).

BR- branch; **CH**- chamber; **EPA**- excurrent papilla; **ICC**- interchamberal canals; **ICO**- interchamberal opening; **ICS**- interchamberal septa; **IPA**- incurrent papilla

FIGURE 16

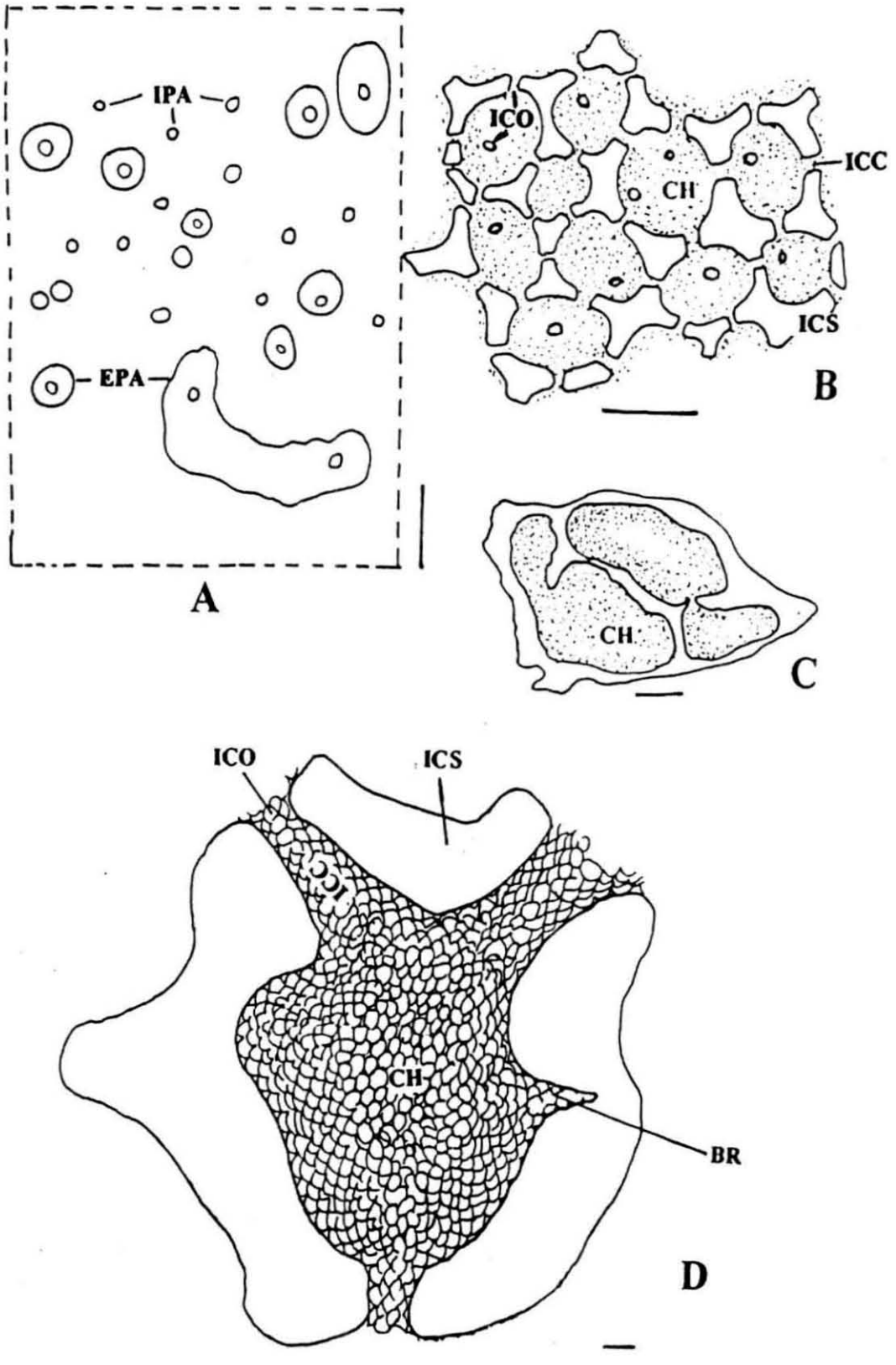


Fig. 17

Cliona celata Grant

A. Vertical section of the mussel shell showing the proliferation of *C. celata* inside. Since the shell is thin, only one tier of chambers is formed inside. These chambers, in advanced stages become irregular due to heavy etching inside the chambers and canals. Excurrent papillae (EPA) may be seen at the upper surface of the shell for the expulsion of water (Shell No. 11, Enayam) (Scale = 1 mm).

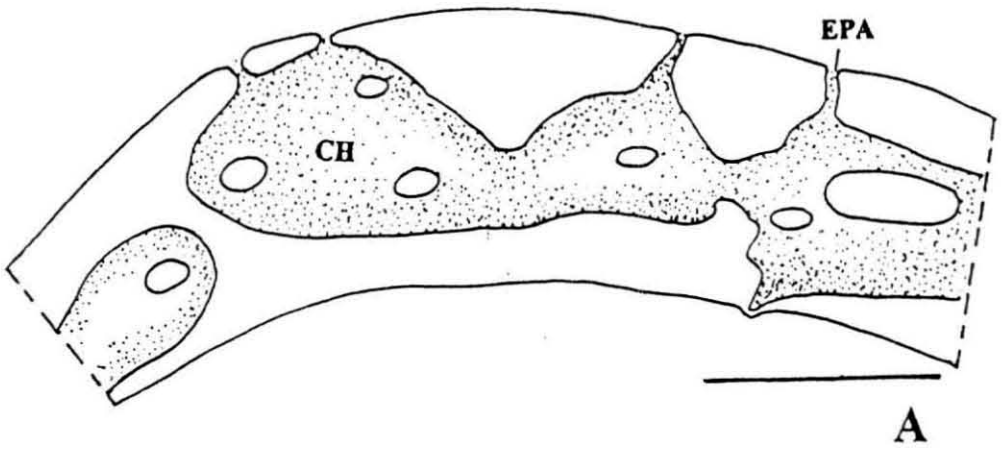
B. Advanced stage of infestation showing extensive tunnel-like cavities formed inside the shell (Shell No. 38, Enayam, Scale = 1 mm).

C. Cavities formed at the thicker parts of the shell. Here the chambers are almost in two tiers and hence the shell is destroyed to the maximum. Periostracum and calcareous layers of the shell are somewhat intact except for the pores through which the papillae project out. Since the middle layers of the shell are excavated to the maximum, slightest pressure would make the shell crumble (Shell No. 48, Enayam, Scale = 1 mm).

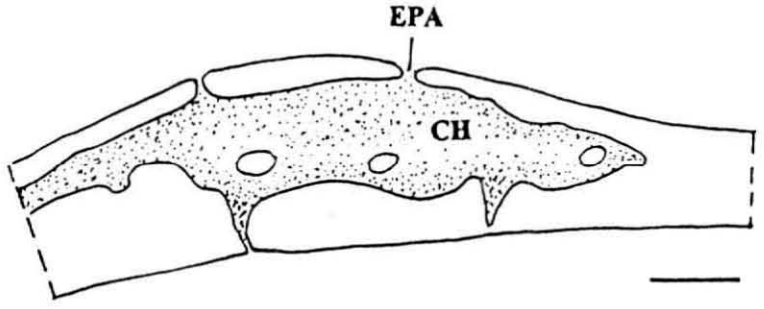
Stippled areas show sponge growth while ustippled areas, the original shell.

CH- chamber; **EPA**- excurrent papilla

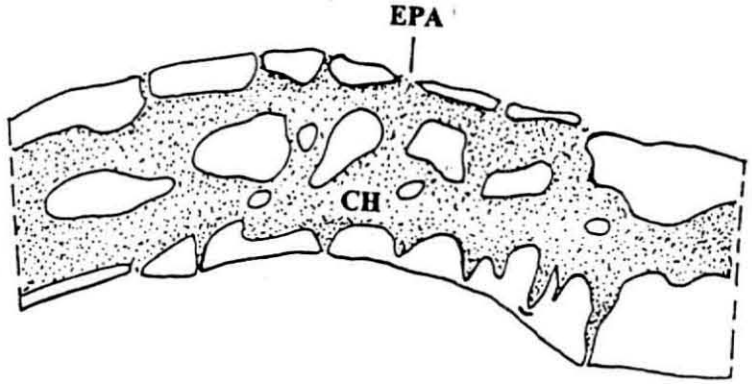
FIGURE 17



A



B



C

Fig. 18

Cliona celata Grant

A. Horizontal section of the shell through LLC (refer Fig. C below for a shell with many tiers of chambers). Since the shell is very thin only one tier of chambers is formed. These chambers are viewed under microscope in the direction of arrow shown in Fig. C from LLC downwards. From the chambers close to nacreous layer it is seen that several papillar projections arise and run through the nacreous layer. In some chambers, only two papillae are seen while in others several are seen (Shell No. 38; Enayam, Scale = 1 mm). Stippled areas represent sponge growth while unstippled, the original shell.

B. One chamber in A is enlarged to show the nature of papillar projections piercing at the nacreous layer. Some of these may open out through the nacreous layer while others, when closed by the secretion of nacreous material by the live mussel, leave a pigment zone at the point of "repair" of the hole excavated by sponge. Since these papillar projections originate from a chamber which is so close to the nacreous layer of the shell and are many in number, they tend to be small and arranged close by. The nacreous layer may become more or less spiny or granulated when viewed from the nacreous layer. Viewed in the direction of arrow pointing downwards shown in Fig. C (Scale = 0.1 mm).

C. Horizontal section of the shell through the lower layer of chambers (LLC). Arrows indicate the angle of examination (Schematic section of a thick shell with many tiers of chambers).

D. The above chamber (B) viewed from the lower surface (in the direction of the arrow marked upwards in C). Some papillae are seen piercing the nacreous layer while others are yet to pierce the nacreous layer. When these openings are closed by nacreous material a black pigment is formed and when such black pigments from adjacent openings merge together, a plate-like pigment is formed (Scale = 0.1 mm). Etching inside the chamber is partly given.

CH- chamber; **LLC**- lower layer of chambers

FIGURE 18

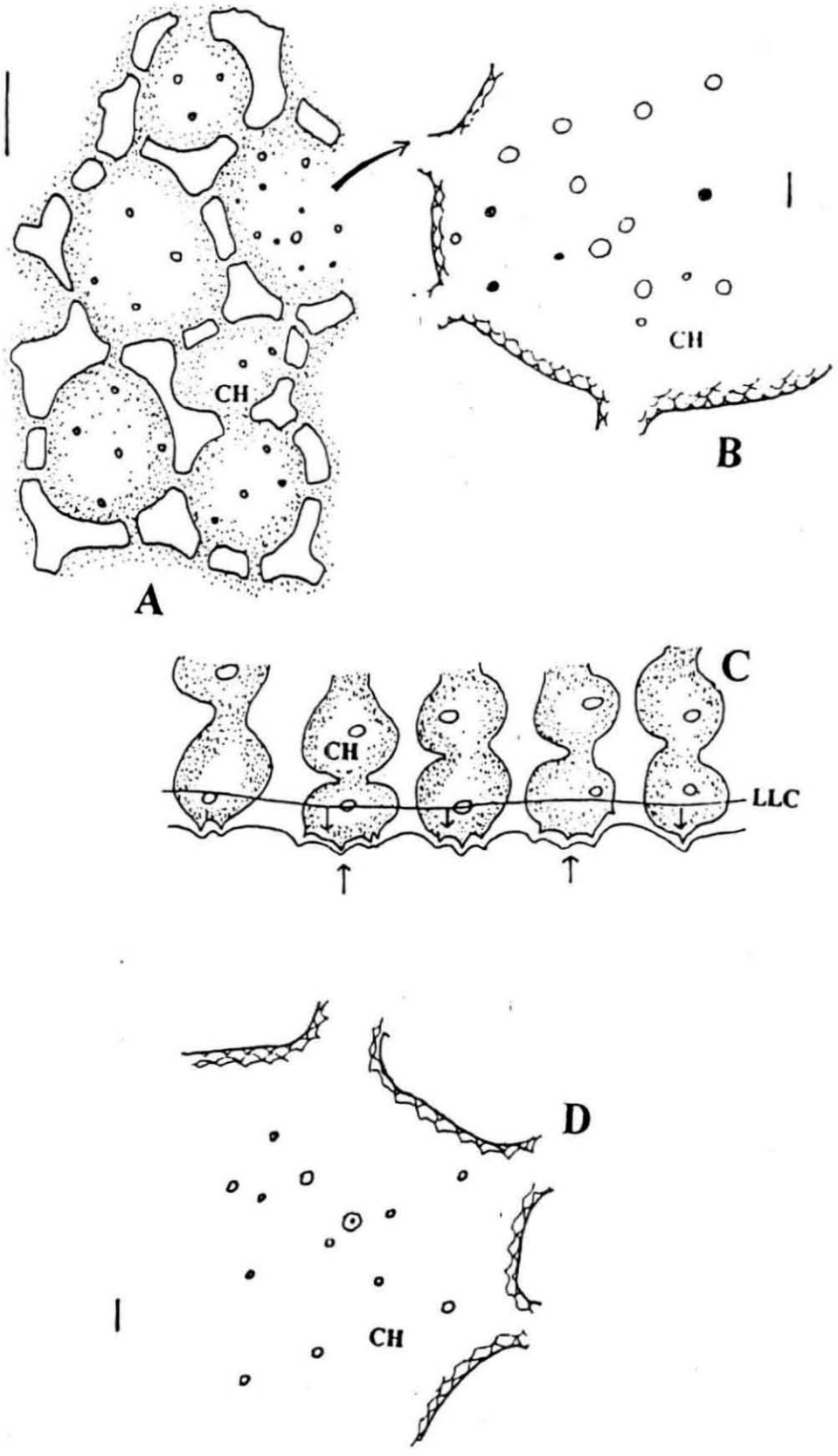


Fig. 19

Cliona celata Grant

Vertical section of the shell showing fully eroded middle layers of the shell. The sponge pierced the inner nacreous layer at many points and the living mussel repaired these openings by secreting nacreous matter. Blisters of different types are produced at the inner side of the shell. Four different types of blisters are seen in the section.

1. Blister with black pigment at the summit.

2. Blister opening out at the submit without any pigment.

3. Blister opening at the summit and with a black pigment ring around the opening.

4. Blister with pigment arranged in a radiating pattern (Shell No. 27, Enayam, Scale = 1 mm).

Stippled areas represent sponge growth, while unstippled, the original shell; 1-4 are not drawn to scale.

CH- chamber; **EPA**- excurrent papilla

FIGURE 19

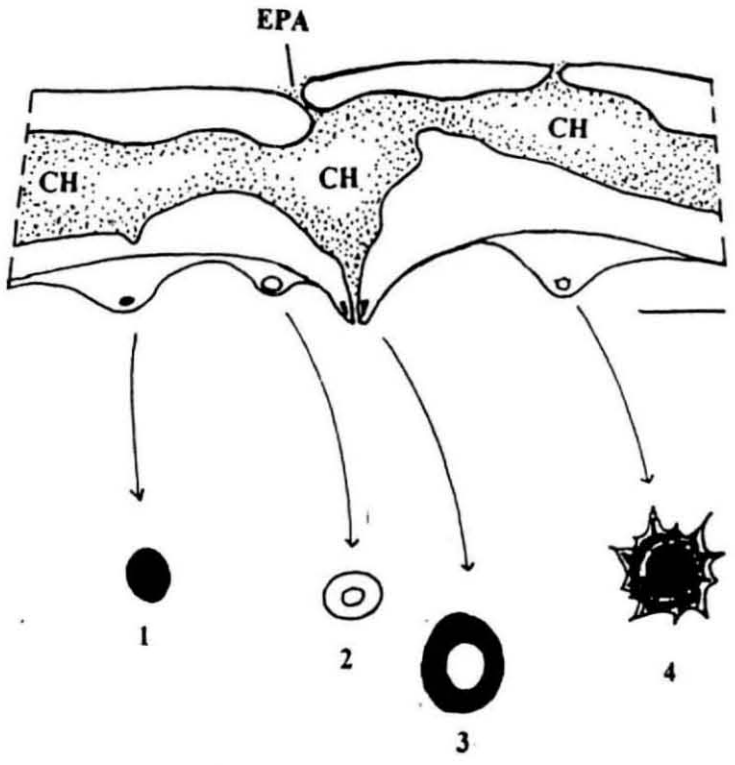


Fig. 20

Cliona celata Grant

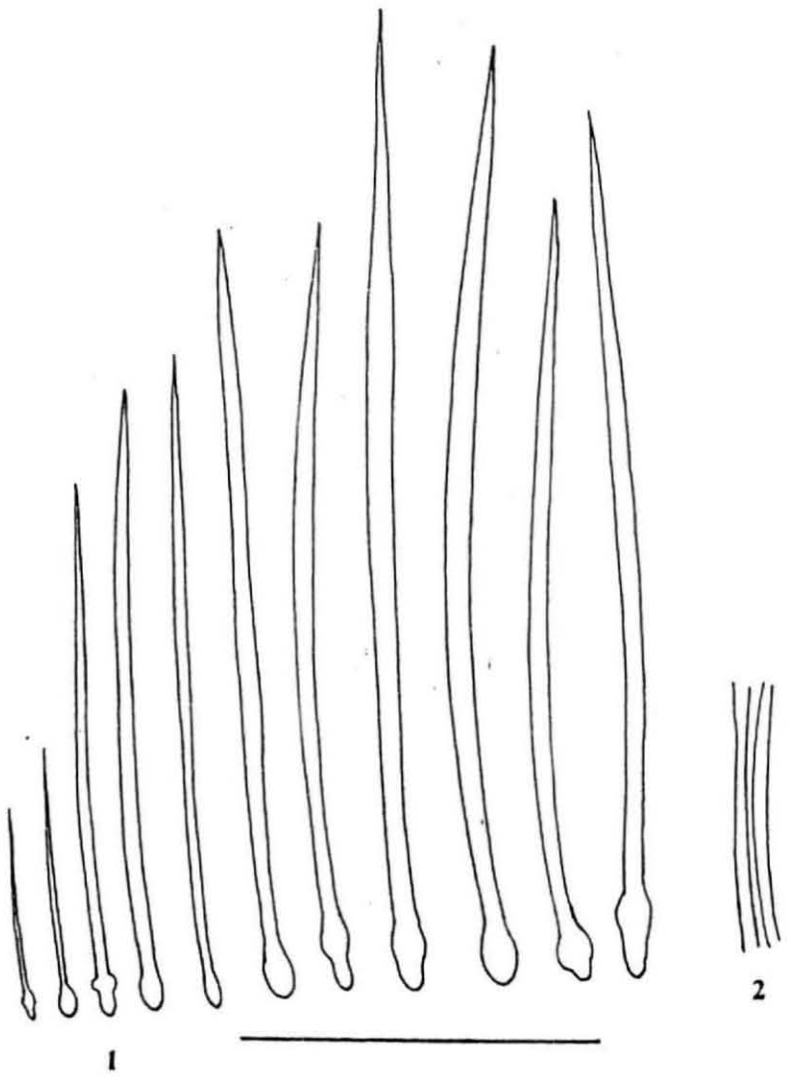
Spicules:

1. Tylostyles: smooth and slightly curved in the first half, tips sharply pointed, head trilobed to oblong.

2. Hair-like oxeas

(Scale = 0.1 mm)

FIGURE 20



3. *Cliona vastifica* Hancock
(Figs. 21-23, Pl. 4 D, Pl. 5 B)

Restricted synonymy:

Cliona vastifica Hancock, 1849, p. 342, pl.15, fig.12; Topsent, 1900, pp. 56-57, pl. 2, figs. 3-9 (synonymy upto 1900); Annandale, 1915 A, p. 8; Annandale, 1915, p. 34, pl. 4, fig. 7; Vosmaer, 1933, pp. 402-411;Old, 1941, p.11, pls. 7& 13; Hopkins, 1956, pp. 44-57; Hartman, 1958, p. 21, fig. 5; Hartman, 1964, p. 3, pl. 1, fig. 5; Thomas, 1972, p. 345, pl.1, figs. 3, 3 A, 3 B (synonymy); Thomas, 1973, p. 61, pl. 3, fig. 11 (synonymy); Rutzler, 1973, p. 633, fig. 6; Thomas, 1979, p. 35, pl. 2, fig.15; Hoshino, 1981, p. 234, fig. 22 (synonymy); Thomas *et al.*, 1983, pp. 1-13; Thomas, 1986, p. 318, pl. 6, fig. 13; Rutzler and Stone, 1986, p. 667; Thomas, 1989, pp. 150-161; Thomas, 1990, pp. 202-216; Thomas *et al.*, 1993, pp. 145-156.

Material: Examined 364 infested brown mussel shells from different Stations (I to VI, Map 1). Apart from these, shells of gastropods and bivalves were also examined from different areas along the coast.

Depth: Mussel beds distributed along this coast in depths varying between 5 m and 8 m.

Colour: Papillae yellow when fresh.

Description: Openings made by the sponge at the surface of the shell are rather irregular at umbo part, while linear and reticulate in the thinner parts of the shell. The diameter of these openings may vary from 0.07 to 0.75 mm; those which accommodate the excurrent papillae (EPA) are larger (0.37 to 0.75 mm), while those which accommodate the incurrent papillae (IPA) are smaller (0.075 to 0.37 mm). In shells of gastropods the diameter of these openings may vary considerably. In advanced stages of boring such openings may be seen on both surfaces of the shell (Fig. 21 A). The incurrent papillae help in taking in water to the interior, while the excurrent one for expelling the water. Both these papillae may project out at the surface to about 2-4 mm in living condition, and contract when taken out of water to the cavity inside the openings. The excurrent papillae are provided with a single opening at the summit (oscule) while the incurrent papillae are with many openings (pores or ostia).

The openings are scattered at the upper surface (umbo part) while they

become linear and reticulate in thinner parts of the shell. In growing parts of the sponge (ie. at the peripheral areas of sponge ramifications) individual branch growing through the middle layers of the shell may be clearly visible. Each branch, after some distance, forms a chamber and then the branch runs in a straight line producing more chambers. From each chamber, thus formed, excurrent and incurrent papillae may be formed and these open out at the surface of the shell. Since the branches of sponge run in a straight line through the middle layers of the shell, the openings formed at the surface also take a linear course and hence these openings, when viewed from outside the shell, give a characteristic linear pattern of arrangement at the surface (Fig. 21 F). In thinner shells, (eg. *Placenta placenta*), the branches and branchlets of sponge formed inside the shell may criss-cross the shell more or less in a straight line giving a reticulate pattern to the surface pores (Thomas, 1983, Plate 1, Fig. D).

The chambers formed inside the shell by the activity of the sponge may be in different tiers in the thicker parts of the shell (umbo proper), while in thinner parts of the shell they may be in one tier running through the middle layers of the shell (Fig. 22 A, B). Inorder to know the structure and size of chambers a horizontal section of the shell was made in the plane ULC and the section was examined in the direction of arrow 3 as given in Fig. 2. It could be noticed that the chambers formed inside are almost oval with a diameter varying from 0.5 to 1.5 mm with interchamberal canals (ICC) and inter chamberal openings (ICO) (Fig. 21 B). The adjacent chambers are separated by the original shell called interchamberal septa (ICS). An examination of the interior of chambers, interchamberal canals and the canals through which the papillae project out at the surface shows a pitted or etched out appearance under high magnification (Fig. 21 C). Diameter of these minute pits may vary from 0.016 mm to 0.07 mm and each pit may represent an area from which a chip has been removed by the activity of the sponge. The interchamberal canal formed by excavating the interchamberal septa (given in box, in Fig. 21 C) is magnified in Fig. 21 E and the interchamberal septa marked in Fig. C (with Δ) is also magnified and given in Fig. 21 D.

As the sponge becomes older and older, the cavities made inside the shell

may get widened due to the removal of calcareous chips from the interior. In more advanced stages the sponge occupies the middle layers of the shell completely making a continuous cavity or tunnel inside. As the volume of sponge increases further, more water may be required for physiological purposes and this may be met by increasing the number of papillae both at the outer and inner surfaces of the shell. The openings made at the nacreous layer may be repaired by secreting nacreous material by the mantle. By constant repair of such openings a blister may be formed and this invariably contain a pigment zone at its summit (Fig. 22 B, B). In some cases the pigment may be seen encircling the papillar canal at its distal extremity (Fig. 22, B, B). The box given in Fig. 22 B is magnified and shown in Fig. 22 C (cross section) and in Fig. 22 D (vertical section.) The pigment zone (PZ) may spread to the interior of the shell to a distance of about 0.15 mm.

The morphology of the species has been well worked out by previous workers (Topsent, 1900; Vosmaer, 1933, and Old, 1941).

Spicules: 1. Tylostyles (Fig. 23 A 1): Straight or slightly curved and sharply pointed. Head spherical when well developed, size. 0.13 to 0.29 x 0.001 to 0.006 mm (length x width), head, 0.005 to 0.008 mm in diameter.

2. Microxeas (Fig. 23 B 2): Microspined in varying degrees or even smooth; swelling may or may not be present at the centre; styloid modifications are rarely noted; size, 0.04 to 0.14 x 0.002 to 0.007 mm (length x width)

3. Spirasters (Fig. 23 B 3): Usually with 3-5 angulations and with spines at angles or entirely microspined; smaller forms with two or three angulations may also be seen, these may be smooth or spiny; size, 0.006 to 0.016 x 0.001 to 0.002 mm.

Remarks: It is widely distributed in the littoral region of the Indian Seas and has even succeeded in colonising the estuarine realms due to its low- salinity tolerance.

Distribution: Cosmopolitan.

Fig. 21

Cliona vastifica Hancock

A. Upper surface (US in Fig. 2, viewed in the direction of arrow 1) of the shell No. 10, (Kadiyapatnam) showing the distribution of pores through which the incurrent and excurrent papillae project out. The oscular openings of the excurrent papillae (EPA) are visible in some (Scale = 1 mm).

B. Horizontal section of the shell (at ULC in Fig. 2, in the direction of arrow marked 3); chambers (CH), interchamberal openings (ICO) and interchamberal septa (ICS) are marked (Scale = 1mm). Stippled areas represent sponge growth, while unstippled, the original shell.

C. One chamber (CH) enlarged to show the etched out-interior (Shell No. 22, Kadiyapatnam) of the chamber (CH), interchamberal openings (ICO) and interchamberal septa (ICS). The initial stage in the formation of inter chamberal canal is marked with a rectangular box and shown in E enlarged, and the end of an interchamberal septa (marked Δ) in D (Scale = 0.1 mm).

D. Area marked with a triangle in the above figure is magnified to show the etched out interior of the chamber (etchings are shown partly only).

E. The inter chamberal septa is getting pierced by the formation of a new branch from the chamber (marked with a rectangular box in Fig. C). Note the etched out interior.

F. Linear growth of sponge inside the shell. Branch (BR) formed from the "sponge mass" inside a chamber runs mostly in a straight line, and later branchlets are formed in a dichotomous pattern. These branches and branchlets bulge out after a short distance to form new chambers. Incurrent papillae and excurrent papillae originate from each chamber and pierce out both at the outer and inner surfaces of the shell (circular markings) for the intake and expulsion of water (Scale = 1mm).

BR- branch; **CH**- chamber; **EPA**- excurrent papilla; **ICO**- interchamberal opening; **ICS**- interchamberal septa; **IPA**- incurrent papilla

FIGURE 21

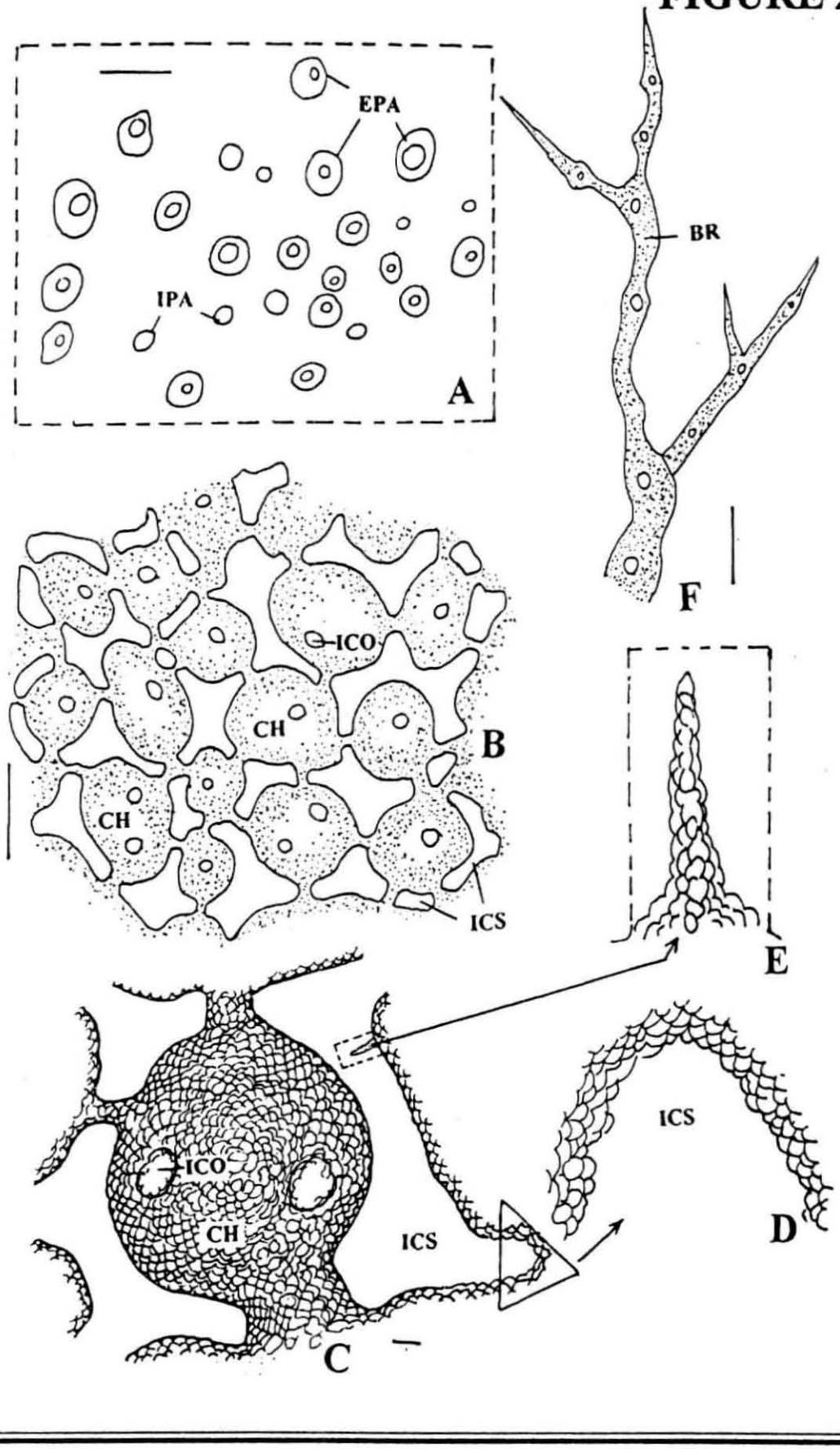


Fig. 22

Cliona vastifica Hancock

A. Vertical section of a mussel shell (Shell No. 18, Enayam) showing the chambers formed inside the shell. Incurrent papillar pores and excurrent papillar pores are shown (IPA and EPA). The sponge already established contact with the soft parts of mussel through papillar openings formed at the nacreous layer (NL). Interchamberal openings (ICO) have attained larger dimensions due to the advanced nature of etching (Scale = 1 mm).

B. Vertical section of mussel shell (Shell No. 13, Kadiyapatnam) showing advanced stage of boring. Individual chambers are not traceable and by fusion of adjacent chambers a tunnel like cavity is formed inside the shell. Two blisters (B) are seen projecting out from the nacreous layer (NL). Here, pigmentation is noted along the distal extremities of papillar canals (PC) somewhat encircling the canals. Section of one blister (in rectangular box) is magnified (in part) and shown in Fig. D and its cross section in Fig. C. (Scale = 1 mm).

C. A cross section of papillar canal and the adjoining pigment zone (viewed LLC downwards as in Fig.2). The inner rim of papillar canal narrows down to papillar openings (POP) at the nacreous layer. From the inner rim of papillar canal (IRPC) the pigment zone spreads to about 0.15 mm into the shell. This zone also narrows down as it reaches the papillar opening (POP) at the nacreous layer (arrow in C indicates the plane of cutting) (Scale = 0.1 mm).

D. A three-dimensional view of the extremity of the papillar canal and the surrounding pigment zone. The papillar canal (PC) narrows down to the papillar opening (POP), which is now closed by nacreous secretion. Pigment zone (PZ) surrounding the papillar canal and the papillar opening are shown. The papillar canal has an etched out interior (Scale = 0.1 mm).

In A & B- stippled areas show sponge growth, while unstippled, the original shell. In C & D the various areas are marked.

B- blister, **CH**- chamber; **EPA**- excurrent papilla; **ICO**- inter chamberal opening; **IPA**- incurrent papilla; **IRPC**- inner rim of papillar canal, **NL**- nacreous layer, **PC**- papillar canal, **POP**- papillar canal opening at nacreous layer, **PZ**- pigment zone

FIGURE 22

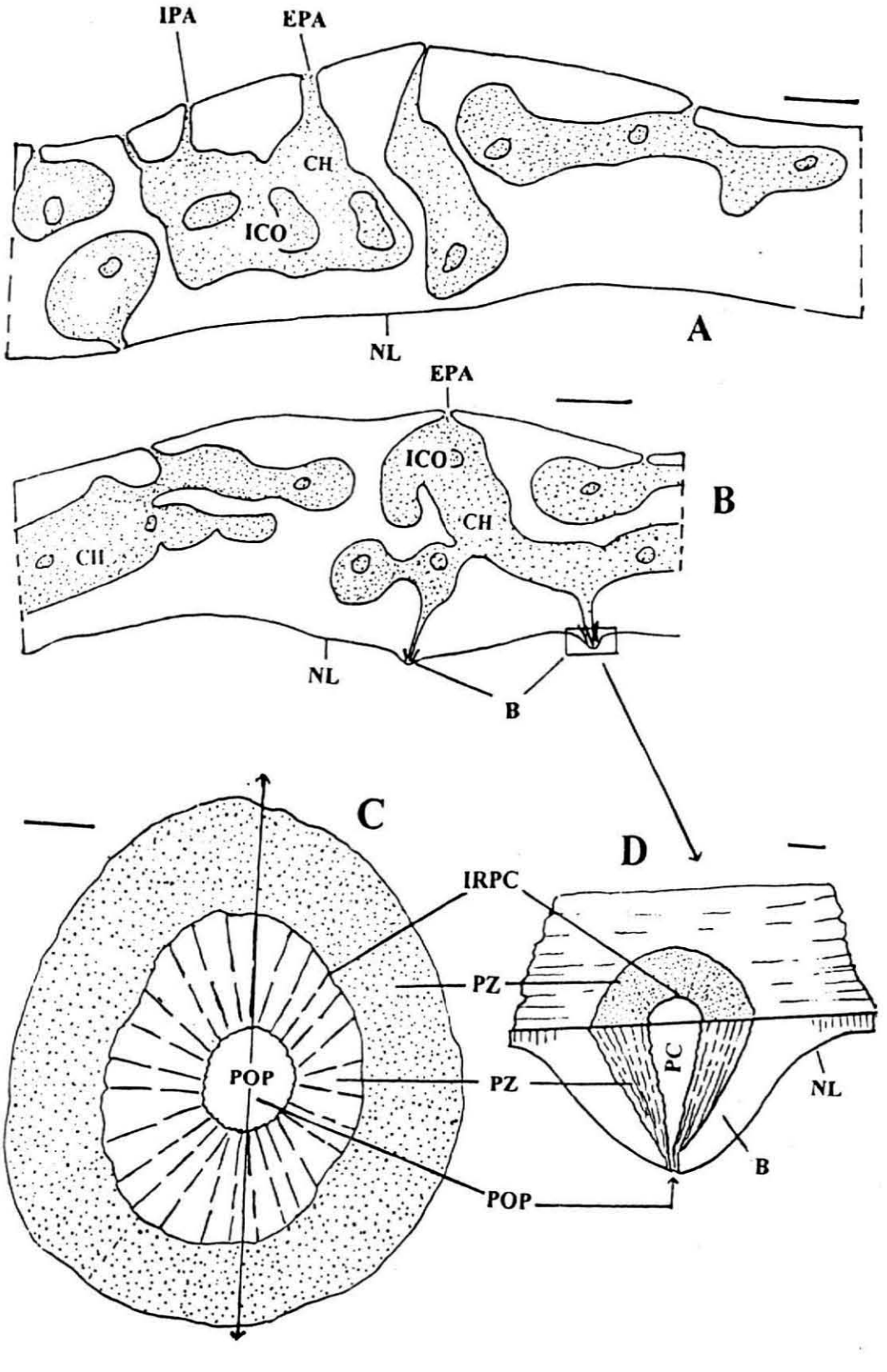


Fig. 23

Cliona vastifica Hancock

Spicules:

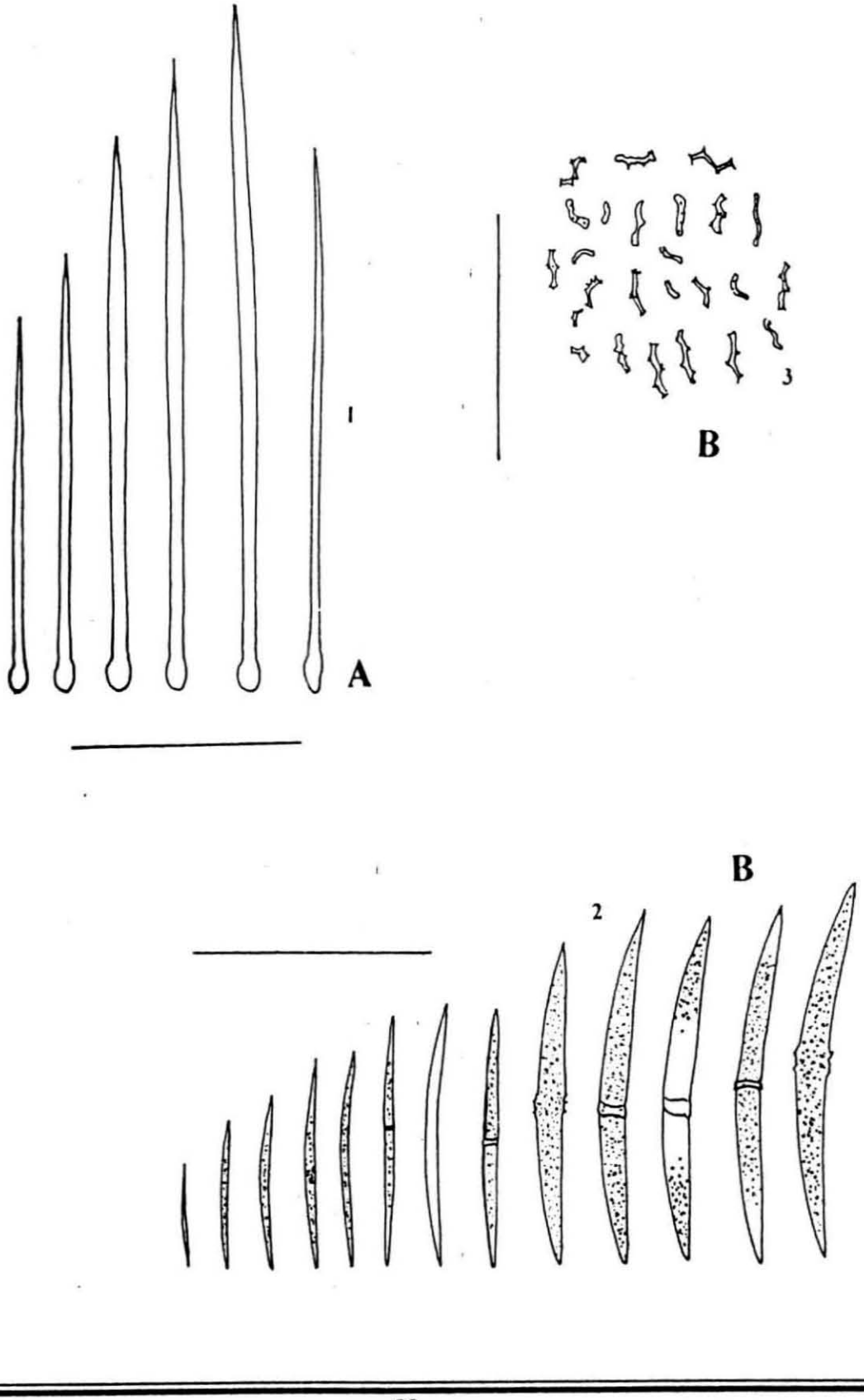
A 1. Tylostyles

B 2. Spined and smooth oxeas

B 3. Spirasters

(Scale = 0.1 mm)

FIGURE 23



4. *Cliona lobata* Hancock
(Figs. 24-27; Pl. 1)

Restricted synonymy:

Cliona lobata Hancock, 1849, p. 341. pl. 12 figs. 4, 8; Topsent, 1900, p. 70, pl. 2, figs. 2, 10, pl. 3, fig.1, pl. 4, fig.1; Burton, 1937, p. 16, pl. 8; fig. 53; Old, 1941, p. 9, pls. 4, 9; de Laubenfels, 1954, p. 215, fig.147; Hartman, 1958, p.19, fig. 4; Thomas, 1979 B, p.172, fig. 2 H; Rutzler and Stone, 1986, p. 663; Thomas *et. al.*, 1983, pp.1-13; Thomas, 1986, p. 318, pl. 6, fig. 14; Thomas, 1990, pp. 202-216; Thomas *et al.*, 1993, pp 145-156.

Material: Examined 443 infested mussel shells from Stations I to VI (Map 1). Apart from the shells of mussels those of gastropods and other bivalves were also examined from the study area.

Depth: Mussel beds in depths varying between 5 and 8 meters.

Colour: Papillae, when extended, are pale yellow to bright red in colour.

Description: Openings made by sponges at the surface of the shell are crowded at umbo part but often in a linear pattern in thinner areas of the shell. The openings through which the excurrent papillae (EPA) emerge out are larger in diameter (0.11 - 0.47 mm) while those through which the incurrent papillae (IPA) project out are smaller (0.037 -0.22 mm) (Fig. 24 A). In advanced stages of boring, such openings may be seen on both surfaces of the shell. The papillae may project out through these openings to a height of about 2-4 mm. Excurrent papillae are provided with a solitary opening (oscule) at their summit, while the pores (ostia) situated at the summit of incurrent papillae are many. Both types of papillae contract when taken out of water and their summits remain flush with the surface of the shell. The oscules may be seen even in the contracted stage.

A vertical section of the shell (Fig. 25 A) shows that the sponge spreads through the middle layers of the shell. The newly formed branches which help in the linear growth of sponge inside the shell are clearly visible at the extreme right hand side of the figure. Further growth of these branches is given in Fig. 25 E (a horizontal view through nacreous layer). Each branch (BR), after a short distance, enlarges to form a

chamber and then the branch continues to grow to form another chamber almost in a straight line. From each chamber both excurrent and incurrent papillae are formed and these open to the outer surface or to the inner surface of the shell. Branchlets may be formed from each chamber and they also function in the same way as the main branch taking almost a straight course inside the shell. Such linear and reticulate pattern of surface openings is seen in *C. vastifica* also (Fig. 21 F).

In order to study the nature, size and minute structure of chambers formed inside the shell, a horizontal section through ULC (as in Fig. 2) was taken and examined in the direction of arrow 3. The chambers formed inside are irregular and the chambers (CH), inter chamberal canals and interchamberal septa are not well defined. Diameter of chambers may vary from 0.8 to 1.8 mm and the inter chamberal canals are more or less ill defined (Fig. 24 B). The interior of chambers (CH) and interchamberal canals (ICC) have an etched out appearance (Fig. 24 C). Each pit represents the area from which a chip has been dislodged by the sponge. The diameter of the pit may vary from 0.029 to 0.076 mm. Chambers remaining close to the nacreous layer were examined for papillar canals and other details (Fig. 24 D), and it could be seen that these are also equally etched out at their inner walls (Fig. 26 B).

In advanced stages of boring the middle layers of the shell get chipped off completely forming a continuous cavity inside rendering the shell quite brittle (Fig. 25 B, C, Fig. 26 A). As more water is required to meet the physiological needs, the sponge may put forth incurrent and excurrent papillae to the inner side of the shell. These papillae are prevented from touching the soft tissue of mantle by secreting additional quantities of nacreous material by the mussel, and by constant repair of such openings blisters may be formed. Pigment formation at the area of repair is the same for all the species described earlier (Fig. 25 D; Fig. 26 B, C). In the case of shells where there is meagre nacreous secretion, the openings made inside the shell may remain open for ever (Fig. 26 D, rock oyster shell with openings inside); diameter of these openings may vary from 0.09 to 0.5 mm).

The morphology of this species has been described in detail by Topsent (1900) and Hartman (1958).

Spicules: 1.Tylostyles (Fig. 27 A): Straight with pointed tips, younger forms may be sinuous; head spherical and with additional swellings near the head; size, 0. 1 to 0.211 x 0.002 mm -0.006 mm, head may have 0.002 - 0.006 mm diameter, additional swellings may have the same diameter as that of the head.

2. Spirasters (Fig. 27 B): They are of different types:

1. long and robust with rarely distributed spines (Fig. 27 B 1); size, 0.127 mm x 0.004 mm (length x width).
2. long and slender forms, which are spirally spined (Fig. 27 B 2); size, 0.12 x 0.001 mm
3. long and robust with spirally arranged spines (Fig. 27 B 3),
4. long zig-zag type with spines arranged at angles (Fig. 27 B 4),
5. long, slender, zig-zag type with spines arranged spirally (Fig. 27 B 5),
6. zig-zag or straight forms which are the smallest of this category (Fig. 27 B 6); size, 0.010 mm x 0.001 mm; some times they may even be smooth.

Remarks: This species is a dreadful oyster pest of the Atlantic, and it is also known from the Gulf of Mannar (Burton, 1937). No other records of this species was obtained from Indian Seas till 1980 when it appeared in the molluscan culture rafts at Vizhinjam (Arabian Sea). Since then it started spreading to gregarious molluscan beds of the Indian Seas and now it forms a sizeable fraction in the species composition in various natural / tended stocks of molluscs.

Distribution: Atlantic and Indo Pacific.

Fig. 24

Cliona lobata Hancock

A. Outer surface of mussel shell (Shell No. 22, Enayam) showing the distribution of pores through which the incurrent and excurrent papillae protrude (IPA & EPA) (US in Fig. 2., viewed in the direction of arrow 1). The oscular opening of the excurrent papilla may be seen in some even in their contracted condition (Scale = 1 mm).

B. Horizontal section of shell (Shell No. 41, Enayam) at ULC (Fig. 2, viewed in the direction of arrow 3). In this case the usual pentagonal pattern of chamber formation is seen only in the initial stages of infestation, but as the growth proceeds the inter chamberal canals and chambers (CH) get widened due to heavy chipping activity at their inner aspect and gradually these canals and chambers become irregular without distinct interchamberal canals and interchamberal septa (Scale = 1 mm). Stippled areas represent sponge growth while unstippled, the original shell.

C. A Chamber enlarged to show the etching pattern (Shell No. 5, Enayam). The inter chamberal canal may be very narrow (see at the bottom of the figure) or very wide (at the left side of the figure) denoting that etching is never uniform in this species. This may give rise to an irregular system of chambers and canals inside the shell in advanced stages of boring (Scale = 0.1mm).

D. Another chamber made inside mussel shell (Shell No. 7, Enayam) (viewed from LLC to nacreous layer as given in Fig. 2 in the direction of arrow 4). Here one interchamberal canals has widened disproportionately. The two smaller interchamberal septa shown at the upper side of the figure have been eroded to the maximum. There is every possibility that these septa may disappear in due course due to heavy boring at these sites. Two papillar canals (PC) directed to the nacreous layer are also shown (Scale = 0.1 mm).

CH- chamber; **EPA**- excurrent papilla; **ICC**- interchamberal canal; **ICO**- interchamberal opening; **ICS**- interchamberal septa; **IPA**- incurrent papilla; **PC**- papillar canals

FIGURE 24

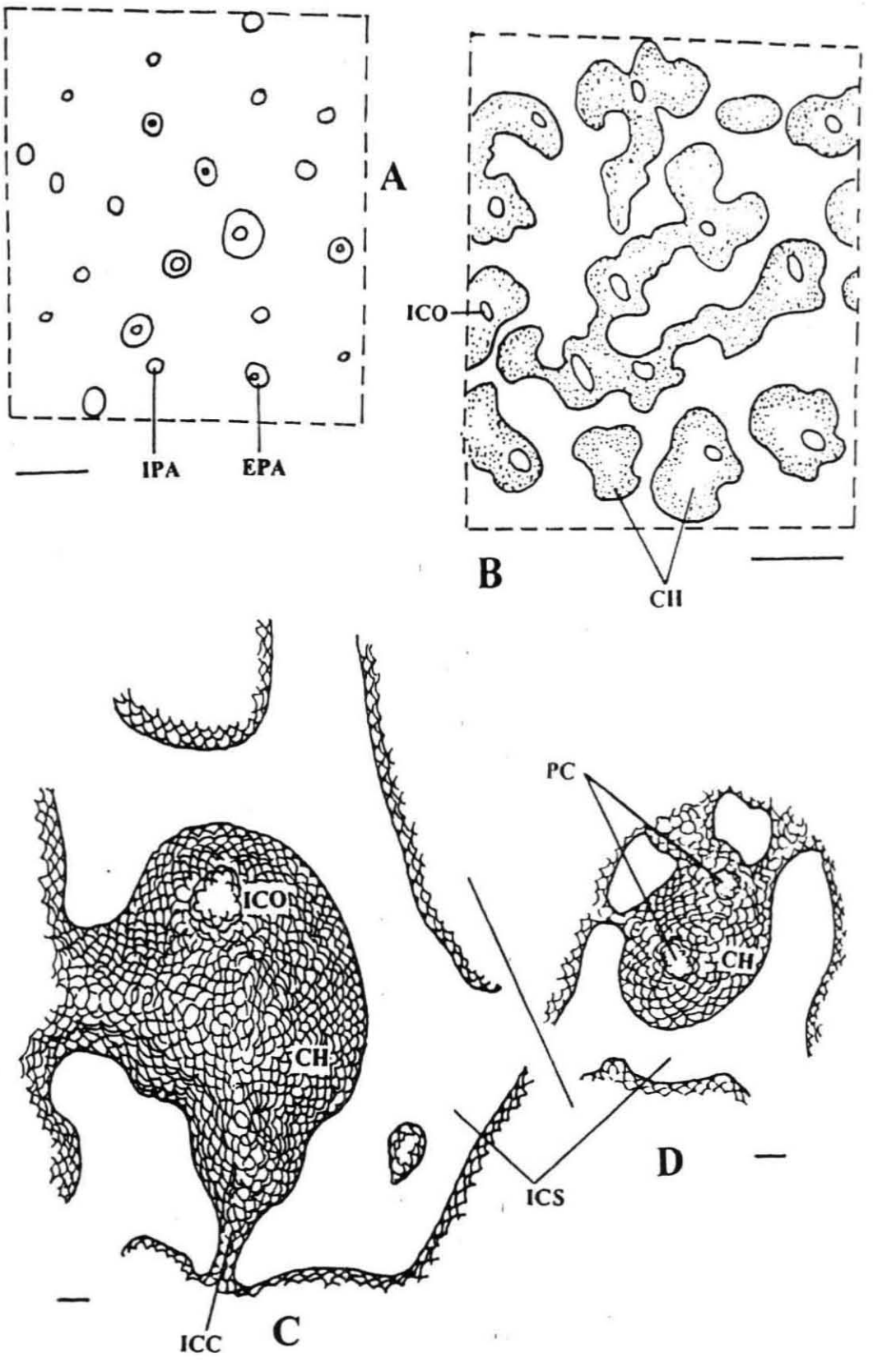


Fig. 25

Cliona lobata Hancock

A. Vertical section of mussel shell (Shell No.16, Colachel) showing extensive cavities formed inside. The interchamberal septa get completely etched out and thus a continuous cavity is formed inside the shell. Branches of sponge proliferating inside are seen clearly at the growing tips (Scale = 1 mm).

B. Vertical section of mussel shell (Shell No. 16, Colachel) with highly eroded interior. Chambers join together and form a wide canal communicating with adjacent canals through wide interchamberal openings. The outer and inner surfaces of the shell are heavily pierced by both incurrent and excurrent papillae (Scale = 1 mm).

C. Vertical section of mussel shell (Shell No. 11, Kadiyapatnam). Here the middle layers of the shell are disintegrated to the maximum. The chambers unite into a continuous cavity in the interior. Many incurrent and excurrent papillae have pierced the nacreous layer establishing contact with the soft parts of the mussel (Scale = 1 mm).

D. Vertical section of mussel shell (Shell No. 34, Enayam) showing the concentration of papillar canals (PC) opening out through nacreous layer. Some of the papillae have pierced the nacreous layer and they were 'repaired' by the mussel (by secreting nacreous material) leaving a distinct black patch (P). One repaired papilla is enlarged to show the details of pigment patch (Scale = 1 mm).

E. Spreading pattern of sponge inside the shell, as seen for the nacreous layer of the shell. Towards the actively growing parts (or the peripheral areas) the branches (BR) are distinctly seen. These branches, as they grow, bulge out to form a chamber and from this chamber papillae may be produced towards the inner and outer surfaces of the shell for the intake and expulsion of water. Branchlets may be formed from each chamber. Branches divide dichotomously and grow almost in a straight line for some distance (Shell No. 16, Colachel; Scale = 1mm).

(In Figs. A to D stippled areas represent sponge growth while unstippled, the original shell).

BR- branch; **CH**- chamber; **PC**- papillar canal; **P**- pigment patch; **PC**- papillar canal

FIGURE 25

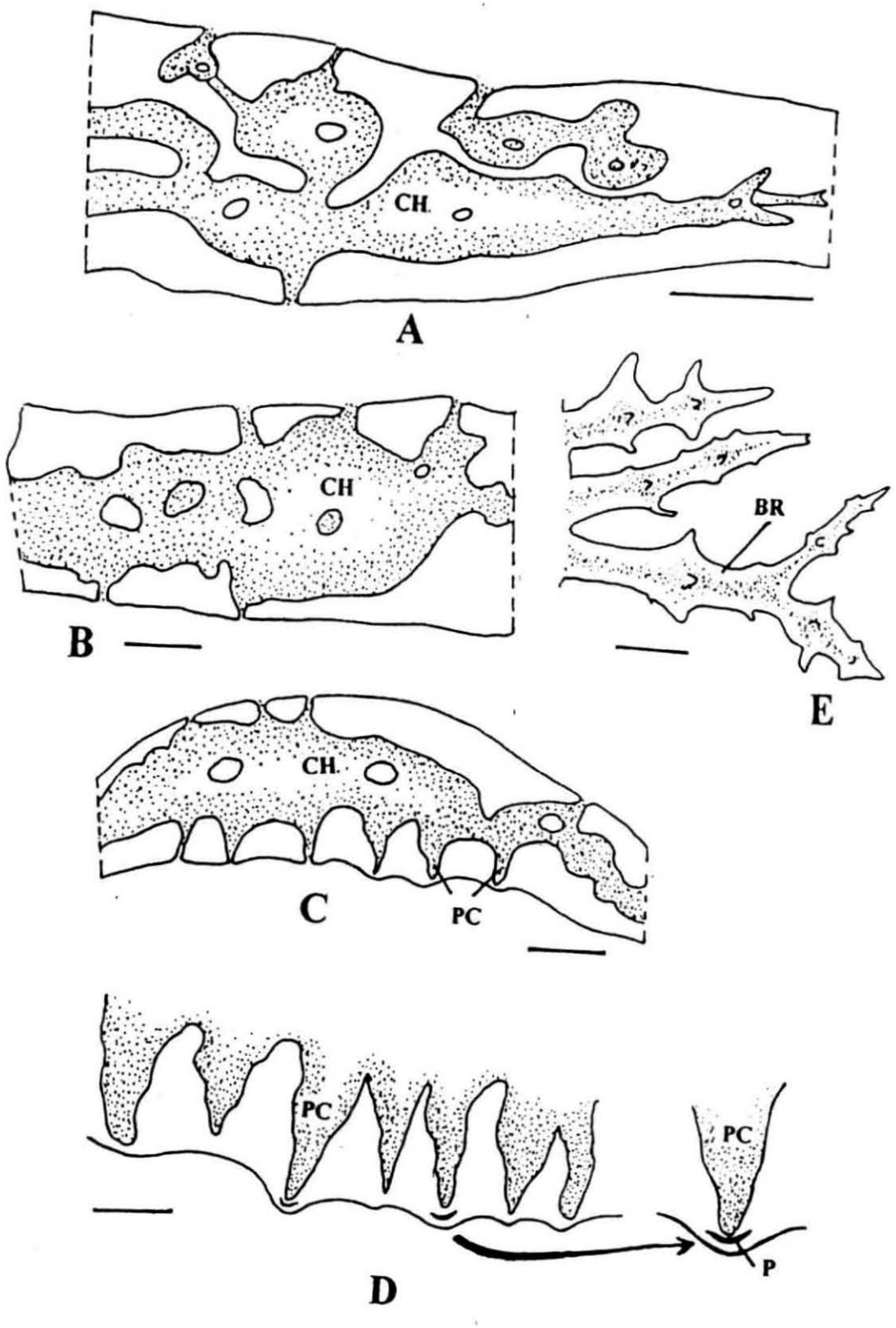


Fig. 26

Cliona lobata Hancock

A. Vertical section of mussel shell (Shell No. 11, Kadiyapatnam) showing heavy damage. Many papillar canals are directed to the nacreous layer (Scale = 1 mm).

B. Magnified view of the papilla marked in Fig. A (in Box). Chamber (CH) and papillary canal (PC) have an etched out interior. Blister (B) formed at the site of "repair" is magnified and given in C with pigment on one side (Scale = 0.1 mm).

C. Blister marked in Fig. B magnified; pigment (P) is partially formed at one side of the blister; repaired hole is also marked (C) (Scale = 0.1 mm).

D. Inner surface of rock oyster shell (No. B3, Vizhinjam) showing the distribution of pores (O). In rock oysters the nacreous secretion is poor and hence the openings made in the inner aspect of the shell remain unrepaired, permitting the contact of papillae with soft parts of the oyster (Scale = 1mm).

B- blister; **C**- closed papillar opening; **CH**- chamber; **EPA**- excurrent papilla; **O**- openings in the nacreous layer; **P**- pigment; **PC**- papillar canal

FIGURE 26

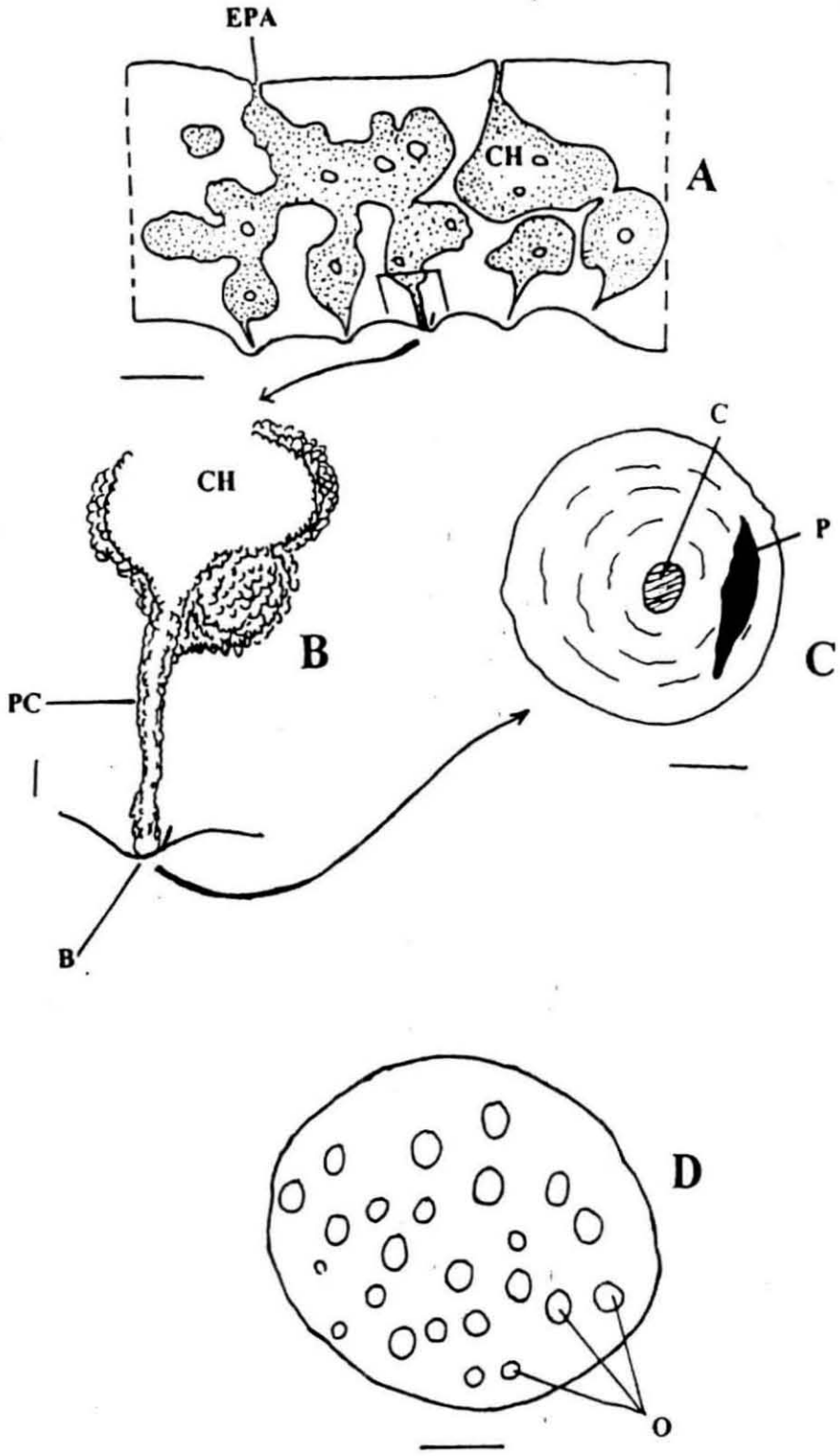


Fig. 27

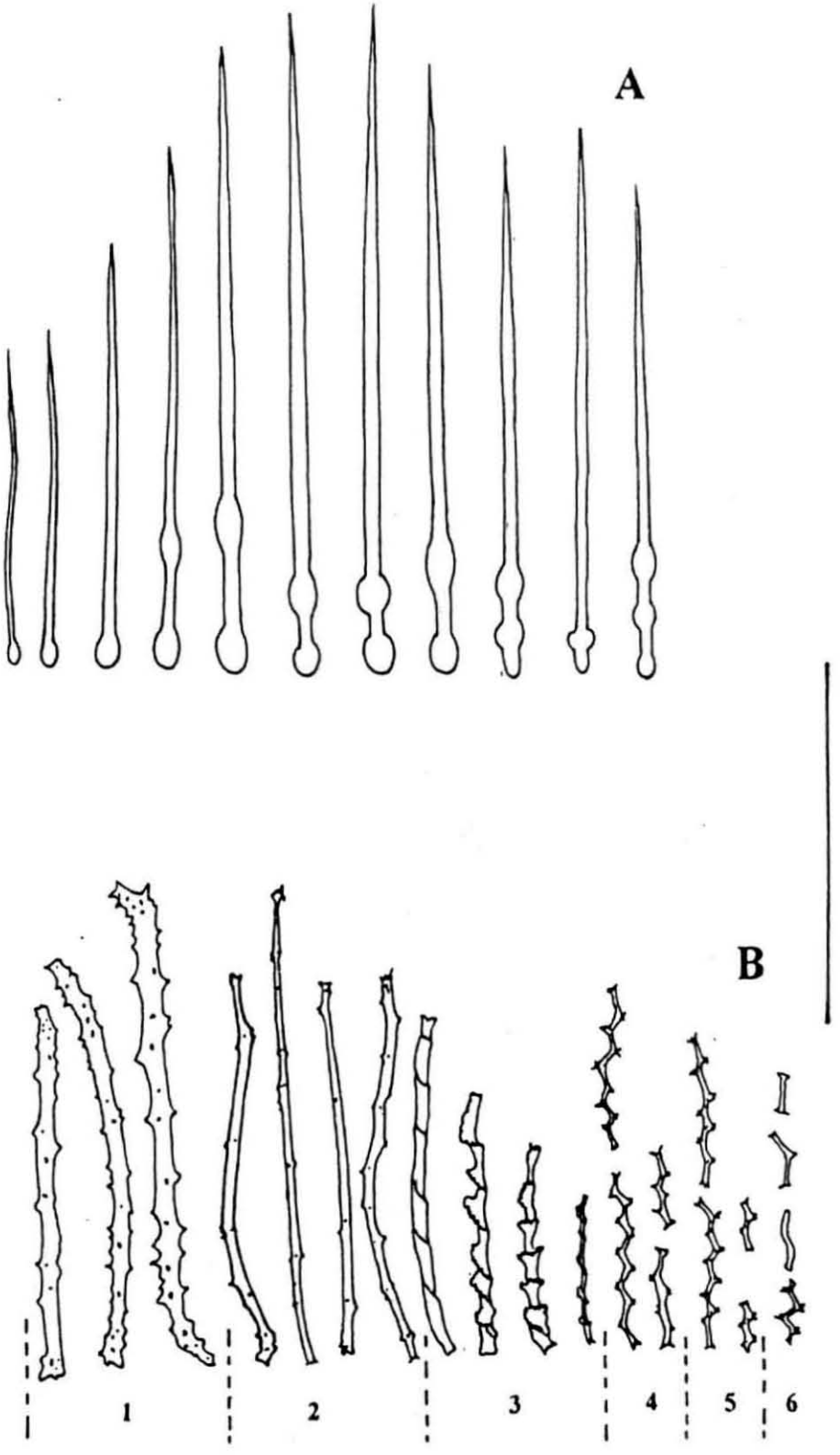
Cliona lobata Hancock

Spicules:

A. Tylostyles: different types

B. Spiraster: different types: 1. Long and robust with rarely distributed spine. 2. Long and slender, spirally spined. 3. Long and robust with spirally arranged spines. 4. Long and slender zig-zag type with spines arranged at angles. 5. Long and slender zig- zag type with spines arranged spirally. 6. Small zig-zag type, with one bent, straight and smooth forms (Scale = 0.1 mm).

FIGURE 27



5. *Cliona carpenteri* Hancock
(Figs. 28- 30)

Restricted synonymy:

Cliona carpenteri Hancock, 1867, p. 241, pl. 8, fig. 4; Topsent, 1888, p. 77, pl. 7, fig. 4; Annandale, 1915 A, p. 8; Annandale, 1915 B, p. 462; Thomas, 1975, p. 122; Thomas *et al.*, 1983, pp.1-13; Rutzler and Stone, 1986, p. 661;Thomas, 1989, pp.150-160; Thomas *et al.*, 1993, pp. 145-153.

Cliona bacillifera Carter, 1887, p. 76

Material: Examined 10 infested mussel shells from different Stations I -VI; (Map 1); shells of other bivalves as also of gastropods were examined from different places along the coast.

Depth: Mussel beds in depths varying between 5 and 8 meters.

Colour: Pale yellow in living condition.

Description: Openings seen at the surface of the shell are circular to oval in outline (Fig. 28 A). In advanced stages of infestation, such openings may be seen in the inner surfaces of the shell (Fig. 29 B). The diameter of these openings may vary from 0.37 to 0.56 mm and the larger openings often, accommodate excurrent papillae (EPA) while the smaller, the incurrent papillae (IPA). Excurrent papillae are provided with solitary opening (oscule) at the summit, while the incurrent pores are many and are arranged like sieve at the summit of the papilla. Usually the terminal part of the papilla is provided with erect tylostyles jetting out in a brush-like pattern in living condition. Both incurrent and excurrent papillae project out at the surface to a height of 2-4 mm in natural condition; they contract into the opening, when disturbed.

A vertical section (Fig. 29 A) of the shell shows that the chambers formed inside are in one tier at the thinner parts while in 2-3 tiers at the umbo part (thicker parts). The sponge inside the shell gets water through the incurrent papillae (IPA) and is expelled through the excurrent papillae (EPA) (Fig. 28 A). The sponge also makes attempt to open its papillae into the inner side of the shell (NL nacreous layer, Fig. 29 A), but the holes made by these papillae are unable to open through the nacreous layer as the openings are constantly repaired by the live mollusc by secreting nacreous

material. Such constant repair of holes made by sponge may lead to the formation of blisters (Fig. 29 A, B) and pigments (Fig. 29 A, P) in the nacreous layer (NL) of the shell.

A horizontal section through ULC (as in Fig. 2; ULC downwards in the direction of arrow 3) shows that the chambers formed are irregular in shape and the inter chamberal septa, so characteristic in all other species, are almost continuous and uninterrupted and the interchamberal canals are considerably widened due to heavy chipping. The diameter of chambers may come upto 2 mm (Fig. 28 B). In more advanced stages of boring, the chambers get united as the interchamberal septa disappear fully through chipping activity of sponge. A chamber, with distinct inter chamberal septa was selected, and the same studied under high magnification (Fig. 28 C). The interior of the chamber (CH) and interchamberal canals showed an etched out appearance as in other species of *Cliona*. Pits formed by the etching away of microchips vary in diameter from 0.025 to 0.072 mm.

Details of the morphology of this species are given in Topsent (1888). The morphology of the "sponge mass" found inside a chamber, branching pattern, etc. are given in Fig. 10 C, which is almost the same for all *Cliona* spp.

Spicules: 1. Tylostyles (Fig. 30 A 1): Straight or slightly curved and with globular head, tips sharply pointed; size, 0.144 to 0.261 mm x 0.004 to 0.006 mm (length x width); head, 0.003 to 0.006 mm in diameter.

2. Microxeas (Fig. 30 B 2): Entirely spined or granulated; size, 0. 046 to 0.113 mm x 0.003 to 0.006 mm (length x width).

3. Bacilliform spicules (Fig. 30 B 3): Straight, fusiform or slightly curved at the centre, spines distinct in some, partly granulated or even smooth forms are present; size, 0.012 to 0.020 mm x 0.001 to 0.002 mm (length x width), very rare in some specimens examined.

Distribution: Atlantic Ocean, Indian Ocean, Australian Region and Pacific Ocean.

Fig. 28

Cliona carpenteri Hancock

A. Upper surface (US) of mussel shell (Shell No. 89, Enayam) viewed in the direction of arrow 1 (see Fig. 2) showing the pores through which the incurrent and excurrent (IPA and EPA) papillae protrude in living condition (Scale = 1 mm).

B. Horizontal section through ULC (see Fig. 2, viewed in the direction of arrow 3). Here in advanced stages, the pentagonal nature of the interchamberal septa is much exaggerated by the widening of chambers (CH) and interchamberal openings (ICO) (Shell No. 88; Enayam, Scale = 1mm). Stippled areas represent sponge growth while unstippled, the original shell.

C. A chamber enlarged to show the etching pattern of the interior. Interchamberal septa (ICS), inter chamberal openings (ICO) are marked. Interchamberal canals are very wide in some, while narrow in others showing that etching is maximum along the inter chamberal canals. The interchamberal septa are usually six in number but these may get divided by branches formed from the 'sponge mass' inside the chamber (Shell No. 88, Enayam; Scale = 0.1mm).

CH- chamber; **EPA**- excurrent papilla; **ICS**- interchamberal septa; **ICO**- interchamberal opening; **IPA**- incurrent papilla

FIGURE 28

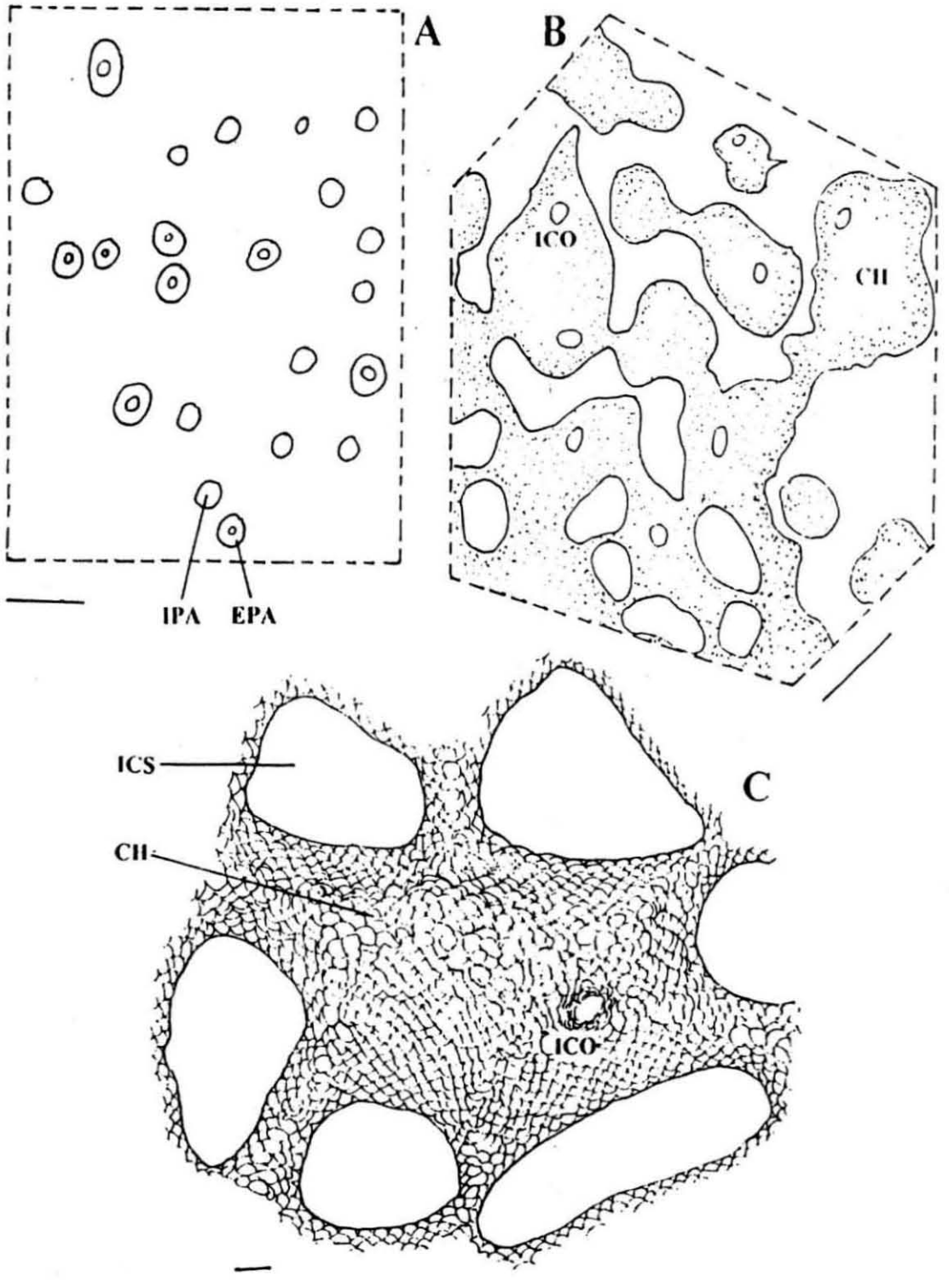


Fig. 29

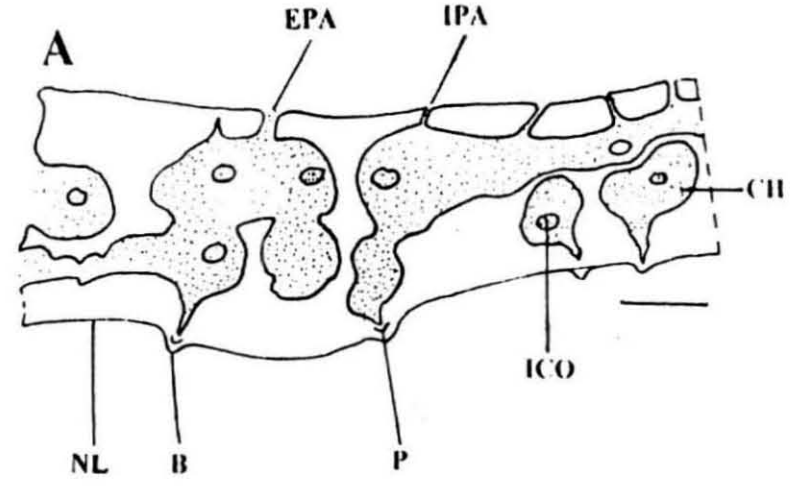
Cliona carpenteri Hancock

A. Vertical section of the mussel shell (Shell No. 29, Enayam) showing the cavities made inside the shell. Here the chambers and interchamberal canals are replaced by a system of tunnels in the middle layers of the shell. Blisters (B), some with pigment (P), are seen in the nacreous layer (NL) (Scale = 1 mm). Stippled areas show sponge growth and unstippled, the original shell.

B. View of the nacreous layer of mussel shell (see Fig. 2, viewed in the direction of arrow 5) showing the incurrent and excurrent papillae (IPA and EPA) in a crowded manner. The excurrent papillae, when contract, a central opening is seen while in incurrent papillae the pores cannot be seen (Shell No. 89, Enayam, Scale = 1 mm).

B- blister; **CH**- chamber; **EPA**- excurrent papilla; **ICO**- interchamberal opening; **IPA**- incurrent papilla; **NL**-nacreous layer; **P**- pigment

FIGURE 29



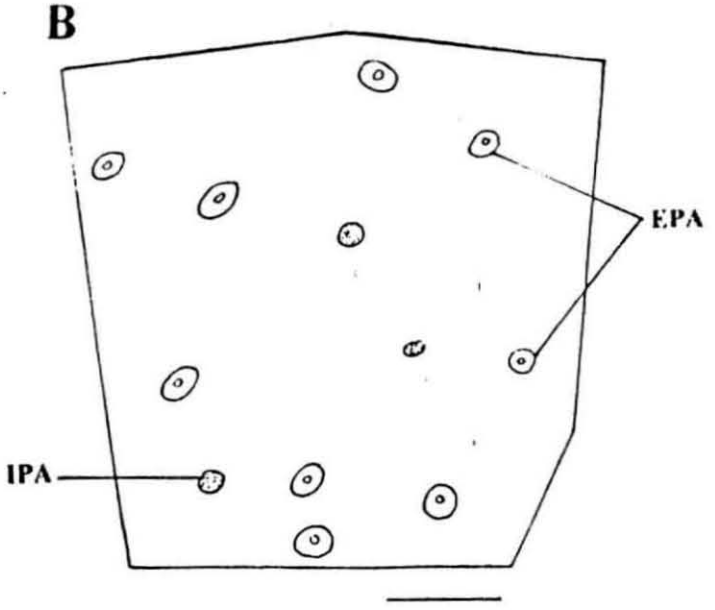


Fig. 30

Cliona carpenteri Hancock

Spicules:

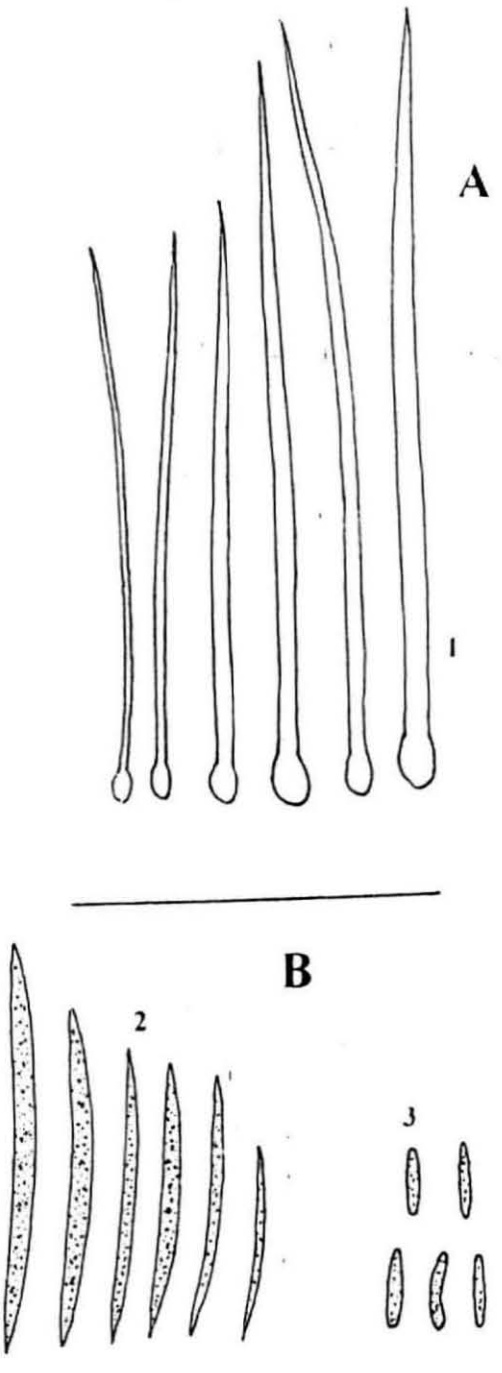
A 1. Tylostyles of different sizes.

B 2. Oxeas, granulated and spined.

B 3. Bacilliform spicules

(Scale = 0.1 mm)

FIGURE 30



6. *Cliona margaritifera* Dendy
(Figs. 31-34)

Restricted synonymy:

Cliona margaritiferae Dendy, 1905, p.128, pl. 5, fig. 9; Hentschel, 1909, p. 386; Annandale, 1915 A, p.9

Cliona margaritifera Vacelet & Vasseur, 1971, p.77, fig. 21; Thomas, 1979, p 36, pl. 2, fig.17; Thomas, 1979 B, p. 177, fig. 5 C; Thomas, 1981, p. 33, pl. 2, fig. 21; Thomas *et al.*, 1983, pp.1-13; Thomas, 1986, p. 321, pl. 6, fig. 21; Thomas *et al.*, 1993, pp. 145-156.

Material: Examined 124 infested brown mussel shells from different Stations (Map 1). Shells of other bivalves and gastropods were also studied from different parts of the southwest coast.

Depth: Mussel beds in depths varying between 5 and 8 meters.

Colour: Papillae yellow in living condition.

Description: Openings made at the surface of the shell, by this species, are much crowded at umbo part. The diameter of these openings may vary from 0.037 to 0.8 mm and those openings with larger diameter often accommodate the excurrent papillae (EPA) while the smaller ones, the incurrent papillae (IPA). These papillae may project out of the surface to a length of 3-5 mm, but when disturbed, they may contract into the respective openings. As in other species of *Cliona* here also the excurrent papilla is provided with a solitary opening at its summit, while the incurrent papilla bears many smaller openings; the summit of the papilla is often protected with tylostyles in a brush-like pattern. When contract, the papillae may occupy a position flush with the surface of the shell. The oscular openings may be seen in contracted condition also when viewed from above (Fig. 31 A).

A vertical section of the shell shows that the chambers made inside the shell by sponge are in one tier with chambers (CH) and interchamberal canals (ICC) progressing almost in a straight line (Fig. 32 A). The water taken in through the incurrent papillae (IPA) is expelled through excurrent papillae (EPA) after circulation. The interchamberal opening (ICO) leads to interchamberal canal (ICC) and thence to the next chamber. But in advanced stages of boring, the chambers and interchamberal

canals get disintegrated due to the heavy etching activity of the sponge and thus a continuous canal running through the middle layers of the shell is formed (Figs. 32 B; 33 D). How new branches are formed from the peripheral part of the sponge and how they run through the middle layers of the shell are shown in Figs. 32 C and D, and as these are the same for *C. lobata* and described earlier, such details are not given here (Fig. 25 E).

For details on the size of chambers and their minute structure, a horizontal section of the shell was taken at ULC (Fig. 2, ULC upwards in the direction of arrow 2) and a study of this section (Fig. 31 B) revealed that the chambers formed are circular to oval in outline with a diameter varying from 2 to 3 mm. Each chamber communicates with the outer side of the shell through two types of openings, the larger one (EPA) lodging excurrent papilla and the smaller (IPA) the incurrent papilla. The interchamberal septa (ICS) are clearly visible in between the adjacent chambers (Fig. 31 B, C). Chambers (CH) and interchamberal canals (ICC) have an etched out interior (Fig. 31 C). Each pit represents the area from which a microchip has been removed by the activity of sponge. The diameter of these pits may vary from 0.037 to 0.075 mm.

Though the papillae through which seawater is taken in and expelled are confined to the outer surface of the shell initially, but they may increase in number in advanced stages of boring with preference to the inner surfaces of the shell (Fig. 32 C). Most of these papillae open into the cavity in between the shell and mantle (Fig. 32 C, PC) and these papillae, when expand, may even touch the mantle epithelium of mussel creating pathological manifestations.

Blisters and pigments are formed at the nacreous layer of the shell (Fig. 33 A-D) as in other species. Some papillae may penetrate through the pigment zone of papillae and then open out in advanced stages. When many smaller papillar canals (PC) are given off from a chamber (Figs. 12 A, 31 D, 33 D) 'pointed blisters' are formed at the nacreous layer giving considerable roughness to the nacreous lining (Fig. 12 A). In such cases, the pigments formed at the base of each blister may unite with the

adjacent one forming a plate-like pigment (PPI) as given in Fig. 33 D. Plate-like pigment formation is seen in *Cliona celata* also.

Spicules: 1. Tylostyles (Fig. 34 A 1): Straight or slightly curved, head globular, oblong or even trilobed; size 0.11 to 0.22 x 0.003 to 0.006 (length x width), head 0.004 to 0.008 mm in diameter.

2. Oxeas (Fig. 34 B 2, 3): They may be uniformly curved or with an angle at the centre, in some forms the central portion with "Z" like bent (B 3). This type, by gradual reduction in size, attains the character of typical spiraster (B 4). Oxeas may be spiny or granulated; spines may be longer at the central part; smooth forms are also met with, size: 0.063 x 0.006 mm (maximum for oxeas).

3. Spirasters: (Fig. 34 B 4): Typical spirasters with 2 to 4 bents and with spines at corners; may measure 0.02 x 0.0008 to 0.002 mm.

Remarks: This species was reported from Ceylon (=Sri Lanka) in 1905 by Dendy as a causative agent for the extensive depletion of the pearl oyster beds. In subsequent surveys this species could not be collected from the Indian Seas. It later reappeared on the pearl culture rafts at Vizhinjam in 1980. Since then it has spread to wild stocks of molluscs in and around Vizhinjam and has even spread to the pearl oyster beds off Tuticorin. A continuous and long term monitoring of the activity of this species is required to ascertain whether the destructive phase will again be repeated in the Gulf of Mannar as has happened in the pearl banks off Sri Lanka in 1902.

Distribution: Indo-Australian.

Fig. 31

Cliona margaritifera Dendy

A. Upper surface of mussel shell (Shell No. 69, Enayam; US in Fig. 2) showing the distribution of openings through which the incurrent (IPA) and excurrent papillae (EPA) project out. The excurrent opening (now contracted) could be seen in some cases (Scale = 1 mm).

B. Structure of chambers as seen from ULC upwards (Fig. 2 viewed upwards in the direction of arrow 2). Chambers are mostly oval to elongate. From each chamber two openings (one excurrent and one incurrent) are seen going to the surface (Scale = 1 mm). Stippled areas represent sponge growth, while unstippled, the original shell.

C. Interior of a chamber enlarged to show its etched out nature. Each chamber communicates with the adjacent one through interchamberal canals (ICC) and the original shell is retained in between the chambers as interchamberal septa (ICS). Both chamber and inter chamberal canals have an etched out interior (Shell No. 52, Enayam) (Scale = 1mm).

D. View from the interior of a chamber to the nacreous layer (LLC downwards, as given in Fig. 2). From these chambers many (usually 2-4) papillar canals (PC) originate and run vertically downwards piercing the nacreous layer. These papillae (both incurrent and excurrent) protrude out through the nacreous layer, creating irritation to the mantle of the live mollusc (Shell No. 2; Enayam, Scale = 1 mm). Stippled areas represent sponge growth while unstippled, the original shell.

CH- chamber; **EPA**- excurrent papilla; **ICC**- interchamberal canal; **ICS**- interchamberal septa; **IPA**- incurrent papilla; **PC**- papillar canal

FIGURE 31

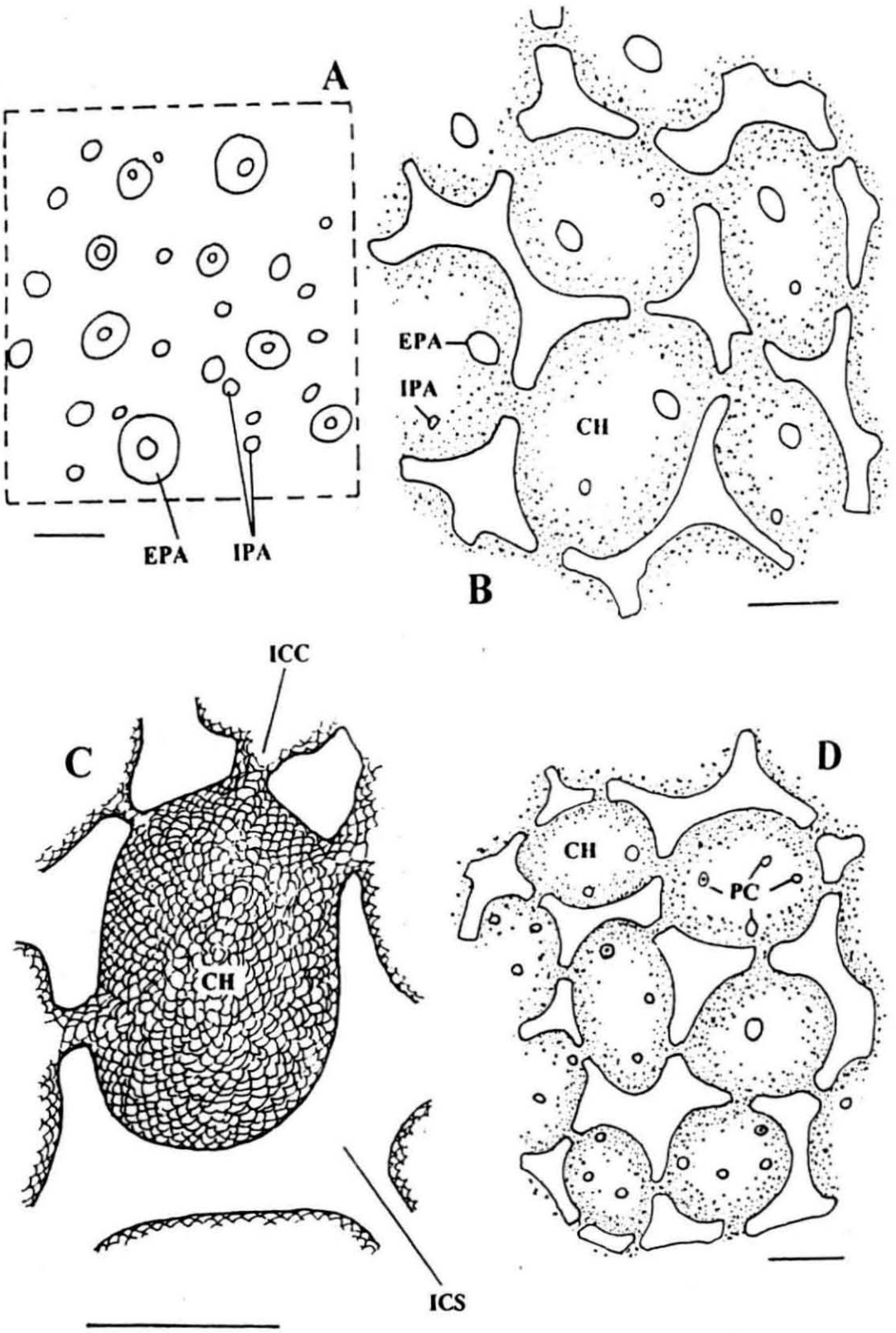


Fig. 32

Cliona margaritifera Dendy

A. Sponge infestation at its initial stages. Chambers (CH) produced inside the shell are spherical and are interconnected with adjacent chambers through narrow inter chamberal connectives (ICC) (Shell No. 6; Enayam, Scale = 1 mm). Stippled areas represent sponge growth, while unstippled, the original shell (vertical section of the shell).

B. Advanced stage of infestation. The chamber - canal arrangement seen in A (above) completely disappears as calcareous particles are chipped off at all sites of sponge contact, and thus a continuous tunnel is formed along the middle layers of the shell. Only the upper and lower layers of shell are intact and such shells, by slightest pressure, will crumble. Stippled areas represent sponge growth, while unstippled, the original shell (Shell No. 69, Enayam, Scale = 1 mm).

C. How branchlets are formed from a chamber containing "sponge mass" is shown in this figure. From a chamber, branchlets (BRL) are formed in a linear fashion and they travel almost through the middle strata of the shell. Such branchlets bifurcate after some distance and thus more branchlets are formed. Vertical section of shell; stippled areas represent sponge growth, while unstippled, the original shell (Shell No. 69; Enayam, Scale = 1 mm).

D. View of branches and branchlets of sponge extending horizontally through the interior. View from nacreous layer in the direction of arrow 5 in Fig. 2. These branchlets (BRL) as they grow, bulge out to form a new chamber and from such chambers branchlets may be formed again and these may make new chambers or pierce the upper and lower surfaces of the shell making incurrent and excurrent papillae (Shell No. 69, Enayam) (Scale = 1 mm). Stippled areas represent sponge growth, while unstippled, the original shell.

BR- branch, **CH**- chamber; **EPA**- excurrent papilla; **ICC**- interchamberal canal; **ICO**- interchamberal opening; **IPA**- incurrent papilla; **NL**- nacreous layer

FIGURE 32

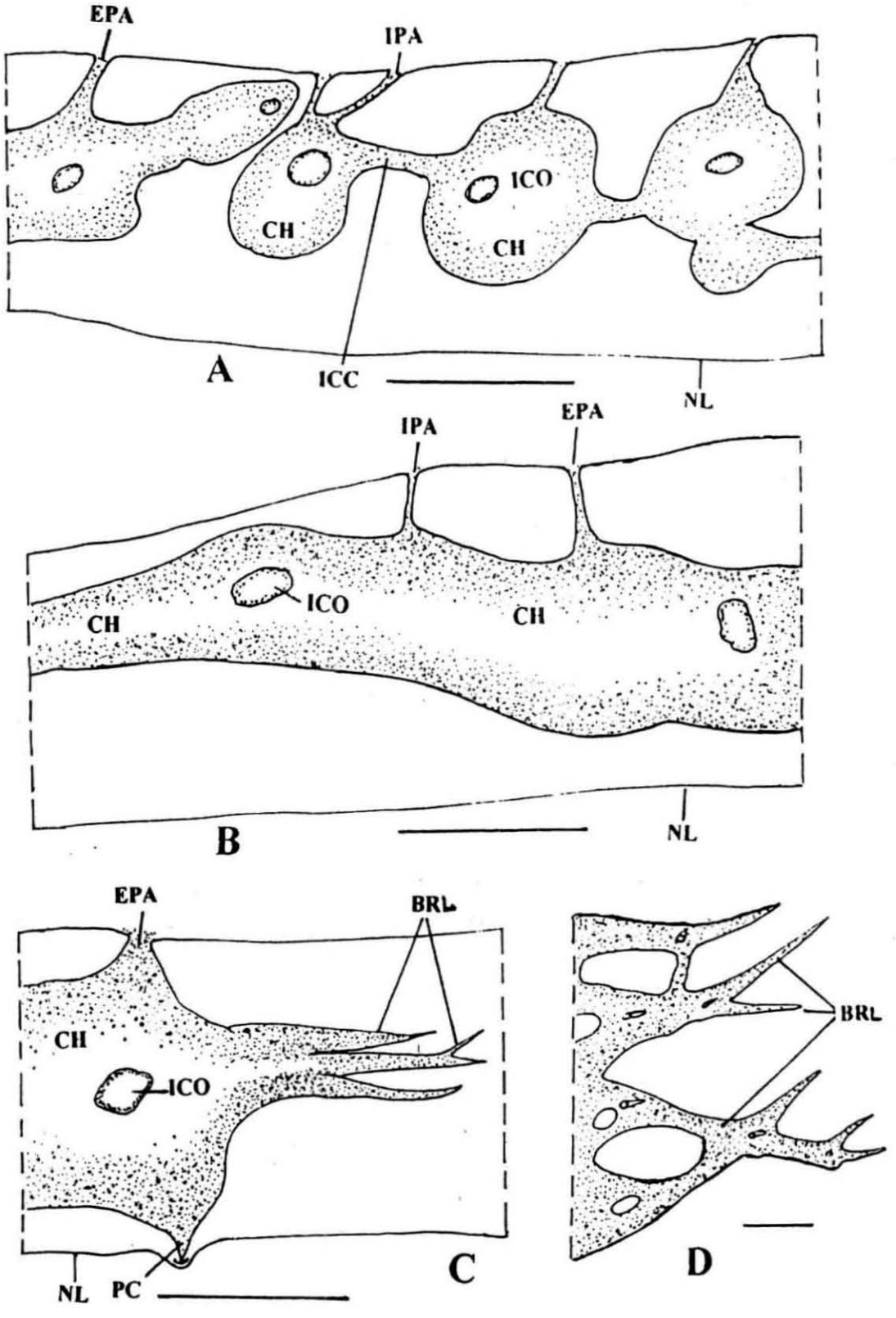

EPA
IPA
ICO
CH
CH
A
ICC
NL
IPA
EPA
ICO
CH
CH
B
NL
EPA
BRL
CH
ICO
C
NL
PC
BRL
D

Fig. 33

Cliona margaritifera Dendy

A. Under surface of the shell (nacreous layer) viewed in the direction of arrow 5 in Fig.2 showing the blisters formed at the nacreous layer due to the boring activity. Blisters are of three types: a. blisters with an opening at the extremity and with out any pigment rim (marked 1); b. summit with a deposition of black pigment and without any opening (marked 2); and c. with opening at the tip of blister and with a black rim (marked 3) (Shell No. 60, Enayam; Scale = 1 mm).

B. Two blisters enlarged to show the black pigment (Shell No. 60, Enayam, Scale = 1 mm).

C. Blister without any opening at the summit. Side view (Shell No. 60, Enayam; Scale = 1 mm).

D. Plate-like pigment (PPI) formation. These are usually produced when chambers (CH) occupy very close to the nacreous layer. From such chambers small papillar canals arise and these pierce the nacreous layer (smaller papillar canals are not shown in figure) producing small granular projections on the nacreous layer. Pigment is formed as and when an opening is repaired. In due course, the pigment, from different such areas (repaired areas) may merge together making a single plate-like pigment (PPI) (Shell No. 60, Enayam; Scale = 1 mm). Stippled areas represent sponge growth while unstippled, the original shell.

CH- chamber; **P**- pigment; **PPI**- Plate-like pigment

FIGURE 33

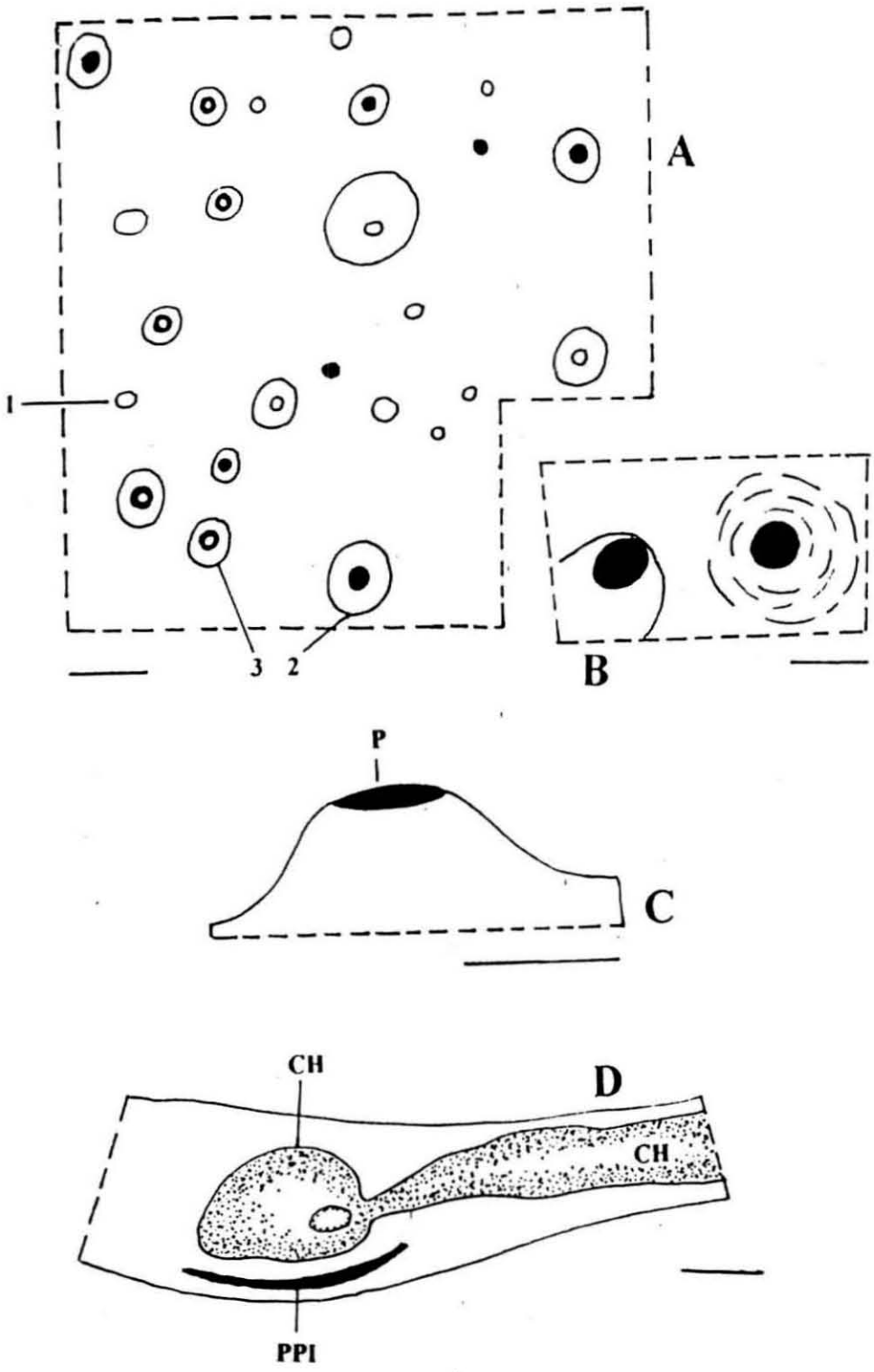


Fig. 34

Cliona margaritifera Dendy

Spicules:

A 1. Tylostyles of different sizes.

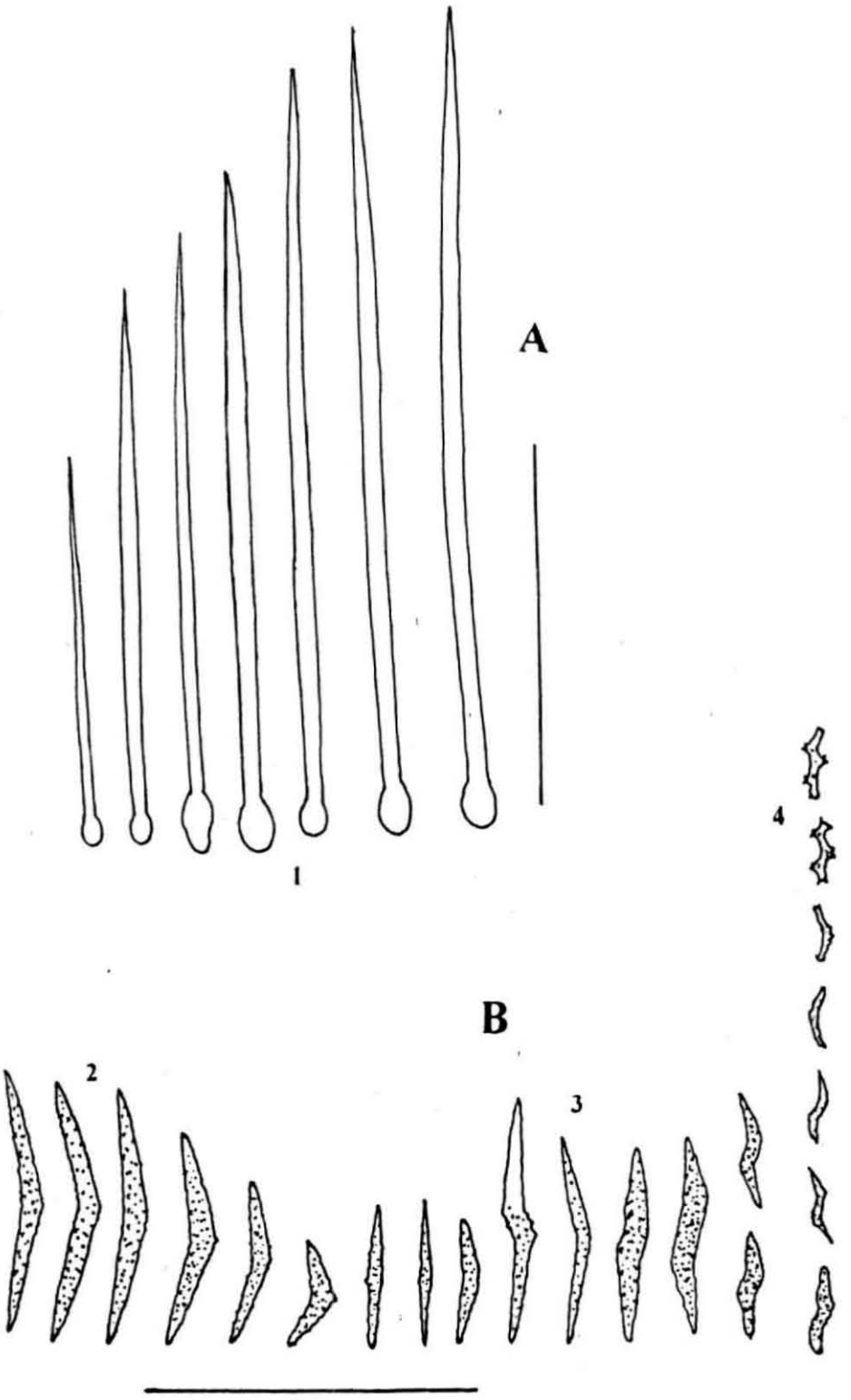
B 2. Oxea, spiny (ordinary type).

B 3. Oxea with a central 'Z' like bent.

B 4. Spirasters (ordinary type).

(Scale = 0.1 mm)

FIGURE 34



7. *Alectona millari* Carter
(Fig. 35)

Restricted synonymy:

Alectona millari Carter 1879 p. 494; de Laubenfels 1936, p. 156; Vacelet, 1969, p. 171; Pulitzer Finali, 1983, p. 500. fig. 29; Bavestrello *et al.*, 1998, pp. 59-70; Carballo *et al.*, 1994, p. 418

Material: Examined two infested mussel shells from Enayam (Station II, Map 1).

Depth: In mussel beds varying from 5 to 8 metres.

Colour: Pale brown when dry.

Description: Only four openings could be seen outside the umbo part in one shell and only two openings in the other. These areas were fully utilised in extracting the spicules, and hence details regarding the chambers, size of chambers, etching pattern, etc. could not be studied in detail. Spicules thus extracted were fully utilised in preparing two permanent slides using Euparol as the mounting medium.

Spicules: 1. Diacts (Fig. 35 A1): Smooth, oxea-like forms with a "kink" at the centre; one or both rays may be wavy in outline; size, 0.126 - 0.260 mm, width upto 0.012 mm.

2. Diacts (robust, Fig. 35 A 2-4)): Entirely spined (weakly or strongly) or tuberculated, tubercles round and mushroom shaped and often supported by a small stalk (magnified and given in Fig. 35, 3 A). Some with an additional arm at the centre (Fig. 35, 4); size, 0.23 - 0.27 mm; tubercles on spicule measure 0.008 mm; width of spicules upto 0.028 mm (inclusive of tubercles).

3. Amphiasters (Fig. 35 B 5 , 6): Fusiform and microspined at tips or not; with two median verticles composed of microspined outgrowths, usually four in number (B 6). Younger forms with conical rays (B 5); size, 0.016 to 0.042 mm (total length).

Remarks: This is the first record of this species from the Indian seas.

Distribution: Mediterranean to North Atlantic and is here recorded from Indian Ocean (Enayam, Arabian Sea).

Fig. 35

Alectona millari Carter

Spicules:

A 1. Diacts, smooth, oxea- like forms with a "kink" at the centre (Scale = 0.1 mm).

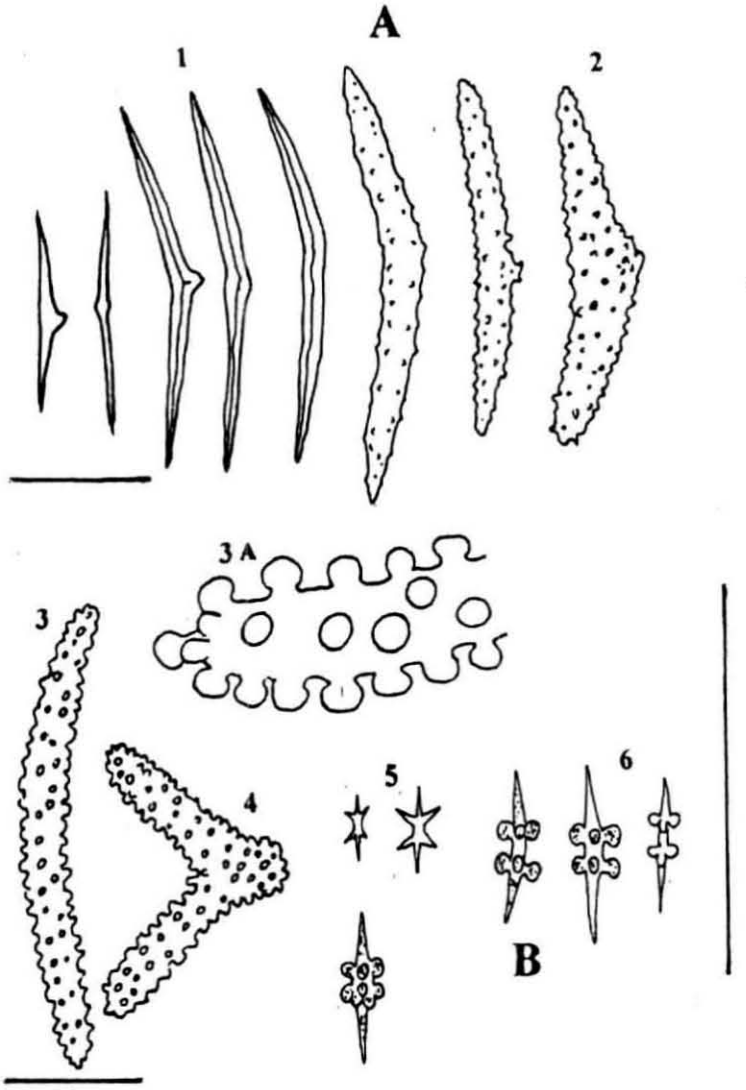
A 2-3. Diacts robust, entirely spined (weakly or strongly) or tuberculated, 3 A, magnified view.

A 4. Diacts with an additional arm at the centre.

B 5. Amphiasters, younger forms with conical rays.

B 6. Amphiasters, fusiform with microspined tips.

FIGURE 35



8. *Thoosa hancocki* Topsent
(Figs. 36-38)

Restricted synonymy:

Thoosa hancocki Topsent, 1888, p. 81, pl.7, fig. 12; Topsent, 1891, pp. 577- 580; Lindgren, 1898, p. 38; Thomas, 1979, p. 37, pl. 2, fig. 18 (synonymy); Thomas, 1979 A, p. 64, pl. 3, fig. 20;

Cliothoosa hancocki Annandale 1915 A, p. 21, Levi, 1965, p. 13, fig. 12; Vacelet, Vasseur and Levi, 1976, p. 40, fig. 21; Rutzler, 1973, p. 634, fig. 7; Schonberg, 2000, p. 179, pl. 3, fig. 15, pl. 7, figs. 37-41, pl. 10, figs. 57, 59, 60.

Material: Examined four infested mussel shells from Enayam (Station No. II, Map, 1) and a rock oyster shell from Vizhinjam (Station No. I, Map 1).

Depth: Mussel beds varying from 5 to 8 metres.

Colour: Dark yellow when alive.

Description: Surface of mussel shell with openings varying in diameter from 0.05 to 0.415 mm. The larger ones lodge excurrent papillae (EPA) while the smaller, the incurrent papillae (IPA). Excurrent papillae, when contract, form ridge-like rim at the surface of the shell and the opening (oscule) one per excurrent papilla, is found in the summit of the rim. The incurrent papillae, when contract, form a mat-like structure at the surface of the shell (Fig. 36 A).

The openings made by this species on the surface of rock oyster are larger: upto 3 mm in diameter for excurrent papillae and upto 1 mm for incurrent ones (Fig. 37 A). The papillae, which open at the inner surface of rock oyster shell are smaller, 1-1.5 mm for excurrent papillae, and 0.5-0.8 mm for incurrent papillae (Fig. 37 B).

A horizontal section of mussel shell through ULC (viewed in the direction of arrow 3 in Fig. 2) reveals that the chambers formed inside the shell are almost irregular and the chambers (CH), interchamberal canals (ICC) and interchamberal

openings (ICO) are well defined only at some places. The interior of the chamber is etched out as in *Cliona* spp. (Fig. 36 B). Chambers formed inside the rock oyster shell are from 3-6 mm in greater diameter and show similar boring pattern as seen in mussel shell (Fig. 37 C, D), but interchamberal canals (ICC) are narrower and drawn out, the etchings are conspicuous both inside the chambers and inside inter chamberal canals (Fig. 37 E, F). The diameter of the pits formed by the removal of chips varied from 0.037 to 0.063 mm.

The nacreous layer of the rock oyster shell possessed several stellate pigment marks (Fig. 37 B, P) but they are not produced as a result of the closure of any opening made by papillae in the inner nacreous layer of the shell.

Spicules: 1. Tylostyles (Fig. 38 A 1): Straight or slightly curved, head oval to rounded, sometimes with an unusual swelling near the head; size 0.21- 0.442 mm x 0.01 - 0.021 mm, head 0.006 - 0.021 mm in diameter.

2. Slender amphiasters (Fig. 38 B 2): Rays long and with recurved terminal hooks; size 0.028 mm when well developed; each arm upto 0.012 mm long. Five growth forms are given in the figure.

3. Nodular amphiasters (Fig. 38 B 3): Rays conical, maximum size upto 0.016 mm, very rare.

Remarks: It is here recorded as a pest on brown mussel.

Distribution: Mediterranean Sea, Red sea, Indian Ocean and Western Pacific.

Fig. 36

Thoosa hancocki Topsent

A. Upper surface (US) of mussel shell (Shell No. 28; Enayam, viewed in the direction of arrow 1 in Fig. 2) showing the openings through which the excurrent and incurrent (EPA & IPA) papillae project out. The excurrent papillae, when contract, leave a small opening at the centre, while the incurrent papillae when contract, the spicules which project out from the surface, may give a furry appearance to the summit of papilla (Scale = 1 mm).

B. Horizontal section through upper layer of chambers (ULC, as given in Fig. 2; examined in the direction of arrow 3) showing the ramifying tunnels formed inside the mussel shell. Newly formed chambers retain their spherical shape while the older ones join together to form a system of tunnels inside (Shell No. 28, Enayam, Scale = 1 mm). Stippled areas represent sponge growth while unstippled, the original shell.

CH- chamber; **EPA**- excurrent papilla; **ICC**- interchamberal canal; **ICO**- inter chamberal opening; **IPA**- incurrent papilla

FIGURE 36

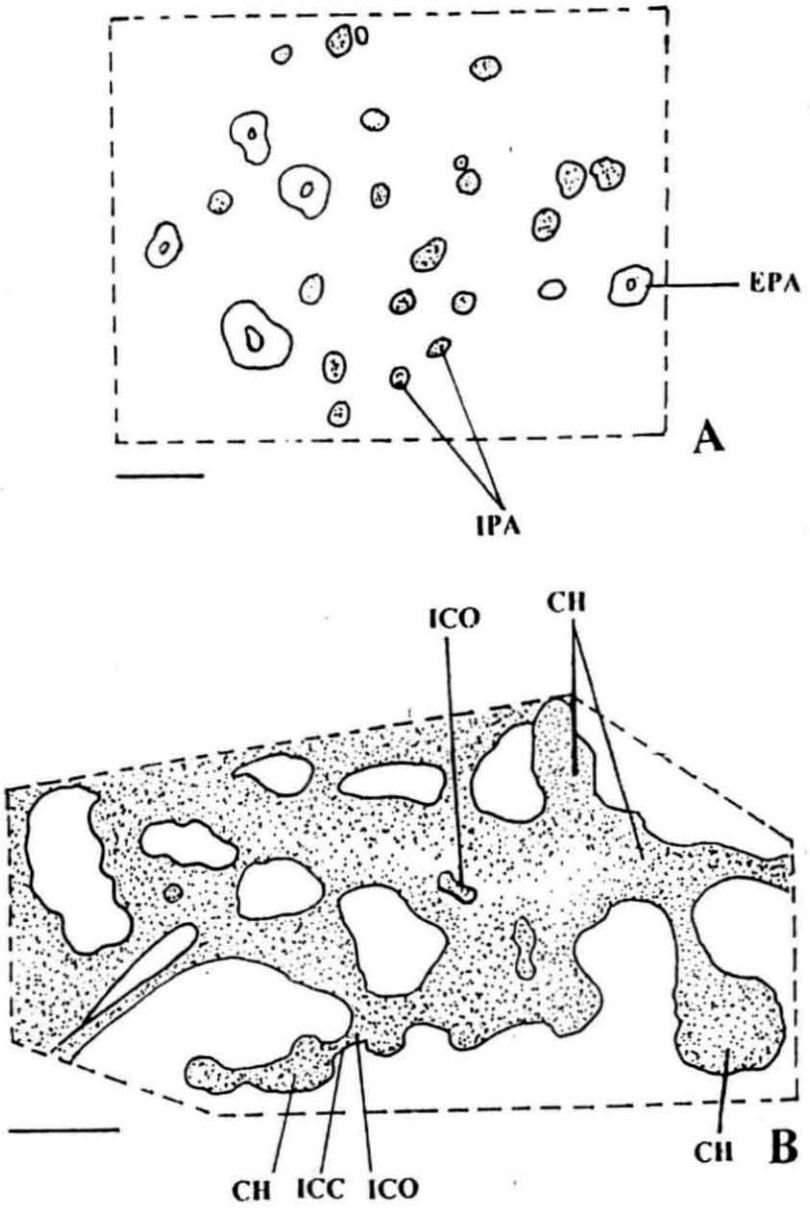


Fig. 37

Thoosa hancocki Topsent

A. Surface of rock oyster shell from Vizhinjam showing the excurrent papillae projecting out from the shell. These papillae, when contract, become thick rimmed and the oscular opening may be clearly visible at the centre (Scale = 1 mm).

B. Inner surface (nacreous layer) of the above rock oyster shell showing the incurrent and excurrent papillae (IPA&EPA). Incurrent papillae are provided with several openings at the summit and when contract these papillae shrink and occupy a position almost flush with the surface while excurrent papillae leave a thick rim outside the surface of the shell. Many pigment marks, often stellate, are also seen embedded in the nacreous layer (Scale = 1 mm).

C. At the initial stage of boring, chambers and interchamberal canals (CH and ICC) are distinct. Rock oyster shell from Vizhinjam (Scale = 1 mm), vertical section.

D. Advanced stage of boring. Here chambers (CH) and inter chamberal canals are no longer distinct. Cavities inside form a system of continuous tunnels. Rock oyster shell from Vizhinjam, horizontal section (Scale = 1mm).

E. Chamber (CH) and inter chamberal canal (ICC) magnified to show the etching pattern (Scale = 0.1 mm).

F. A chamber enlarged to show its etched out interior. Rock oyster shell from Vizhinjam (Scale = 0.1 mm). Etchings are shown partly only; the entire chamber is pitted in the same pattern (In, Figs. C&D- stippled areas show sponge growth and unstippled, the original shell).

CH- chamber; **EPA**- excurrent papilla; **ICC**- interchamberal canal; **ICO**- interchamberal opening; **IPA**- incurrent papilla; **P**- pigment

FIGURE 37

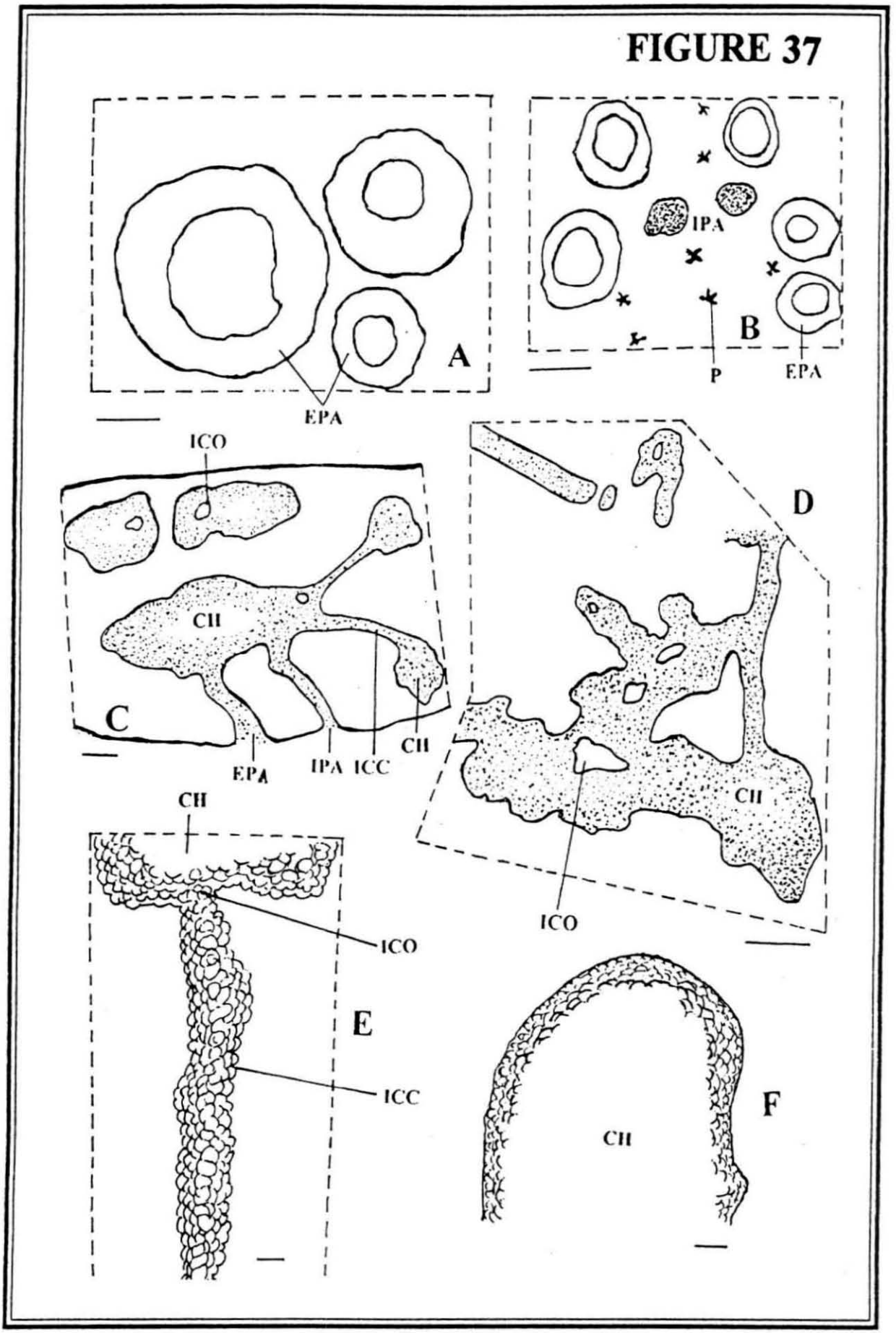


Fig. 38

Thoosa hancocki Topsent

Spicules:

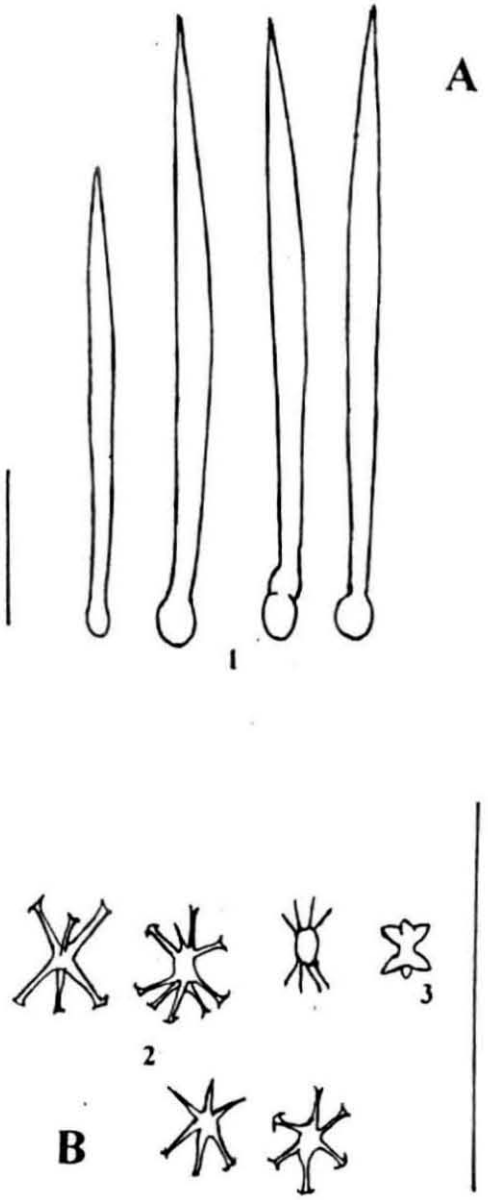
A 1. Tylostyles, different sizes.

B 2. Slender amphiasters.

B 3. Nodular amphiaster

(Scale = 0.1 mm).

FIGURE 38



9.*Thoosa armata* Topsent
(Figs. 39-40)

Restricted synonymy:

Thoosa armata Topsent, 1888, p. 81, pl. 7, fig. 9; Topsent, 1891, p. 579; Topsent 1904, p. 109, pl. 11, fig. 5; Annandale, 1915 A, p. 21; Topsent, 1918, p. 559; Arndt, 1927, p.11, fig. 6; Levi, 1959, p 124; Levi, 1965, p.12; Thomas, 1973 p. 62, pl. 3, fig. 12, pl. 5 fig. 5 (synonymy); Thomas, 1979 B p.18, pl. 181, figs. 2A, 3 B; Thomas, 1989, p.160

Material: One mussel shell infested with this species from Enayam (Station II, Map 1).

Depth: Mussel bed in depths of 5 to 8 meters.

Colour: Papillae pale yellow when dry.

Description: Only three opening outside the umbo part could be located; diameter of opening from 0.5 to 1 mm; they are distributed irregularly. Three papillae could be seen projecting out from the openings and their lateral view is given in Fig. 39 A. The diameter of these papillae varied from 0.3 to 0.4 mm and the entire surface is ornamented with tylostyles projecting from the interior. In one papilla, when examined from above, a central opening (oscule) could be seen and hence this may represent the excurrent papilla while in others no openings could be noted indicating that they are of incurrent nature. Oscular summit, when contract, is protected by tylostyles arranged in a radial pattern with their tips pointing towards the central opening or oscule (Fig. 39 B).

The chambers formed inside the shell are in one tier and the diameter of chamber (CH) may come upto about 0.4 mm (Fig. 39 D). The interior of chambers (CH) and interchamberal canals (ICC) have an etched-out appearance (Fig. 39 C, D) each pit representing the cavity from which a microchip has been removed by the activity of the sponge. The diameter of these pits may vary from 0.025 mm to 0.04 mm.

Spicules: 1.Tylostyles (Fig. 40 A 1): Straight, head globular; size, 0.126 - 0.21 mm x 0.004 - 0.008 mm (length x width); head well developed, 0.008 mm in diameter.

2. Amphiasters (Fig, 40 B 2): Rays in two sets, ray capitate and with microspined head; size: 0.021 x 0.016 mm.

3. Amphiasters with lanceolate rays. Not seen in the present specimen.

4. Amphiasters with long rays (Fig. 40 B,3): those represented in the present specimen may be in their initial stages of development; size, 0.016 mm x 0.012 mm (length x total width, including rays).

5. Oxyasters (Fig. 40 B 4): centrum small; rays long and abruptly pointed; rays may show a reduction in their number; those with two rays are common, but those with three rays are rarely seen; size, for spicules with two rays, 0.06 mm to 0.17 mm, width of a ray from 0.002 mm to 0.003 mm.

6. Oxeas (Fig. 40 B 5): rare, often with a swelling at the centre; rays sharply pointed, length, 0.12 - 0.15 mm, width about 0.002 mm.

Remarks: This is the first record of this species from a mussel shell.

Distribution: Tropical Atlantic, Red Sea and Indian Ocean.

Fig. 39

Thoosa armata Topsent

A. Side view of partially contracted papillae (incurrent and excurrent) projecting out at the surface of mussel shell (Shell No. 28, Enayam) (Scale = 0.1 mm).

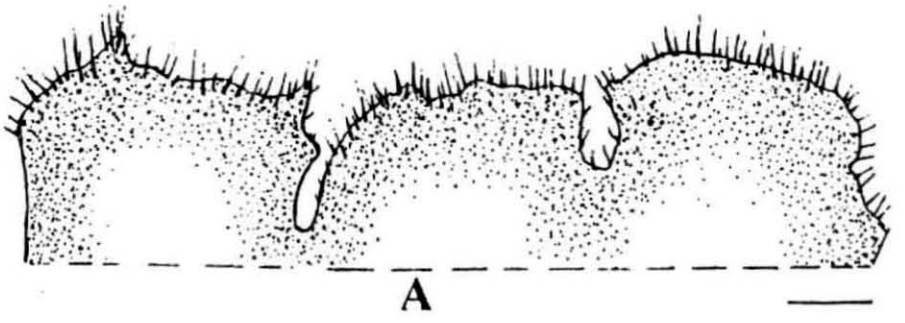
B. Excurrent papilla viewed from above showing the oscular opening. This papilla, when contract, the spicules direct to the central opening arranged radially. Only few spicules are shown in the figure (Scale = 1 mm).

C. Cross section of the interchamberal canal (ICC) showing the etched out interior (Scale = 0.1 mm).

D. Part of the chamber (CH) with interchamberal septa (ICS) showing the etched out interior of the chambers. Etchings partly shown (Scale = 0.1 mm).

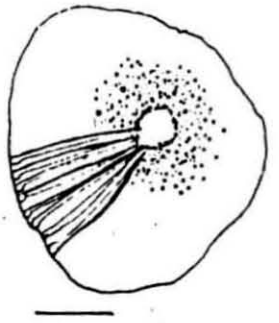
CH- chamber; **ICC**- interchamberal canal;
ICS- interchamberal septa

FIGURE 39

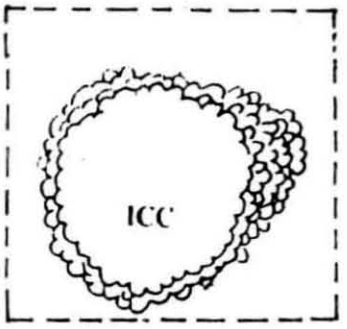


A

B



C



D

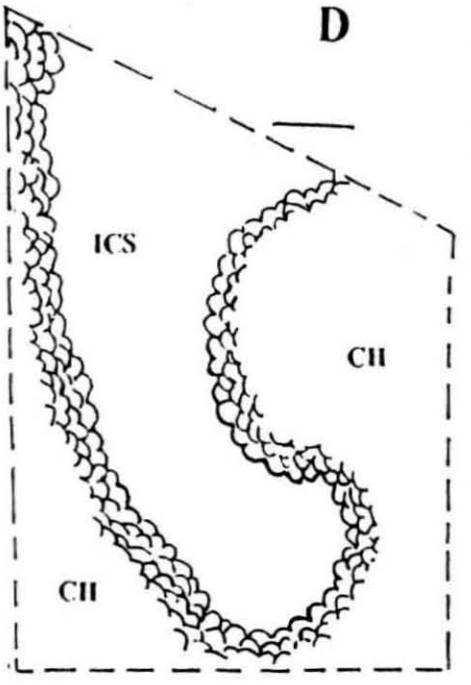


Fig. 40

Thoosa armata Topsent

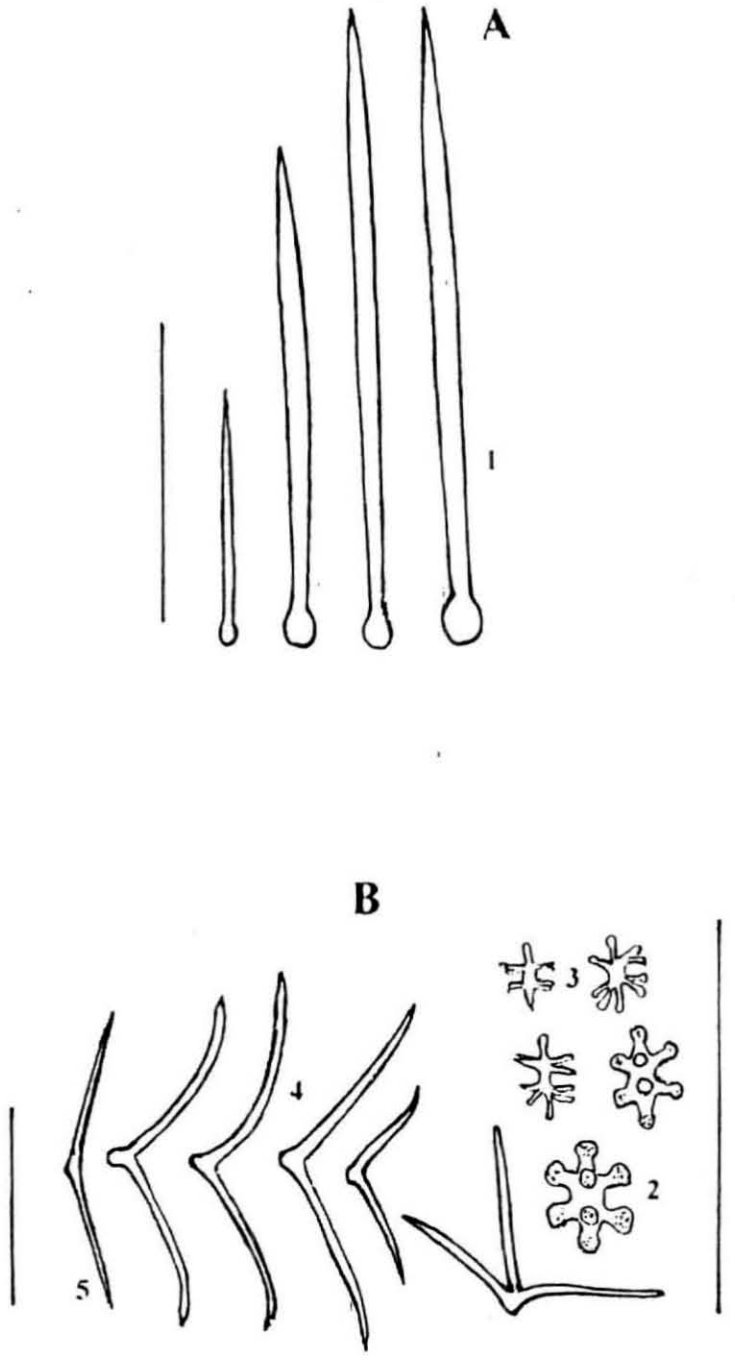
Spicules:

A 1. Tylostyle- different sizes.

B 2. Amphiaster, rays with microspined heads; B. 3. Amphiaster, rays slender and pointed to blunt; B. 4. Oxyaster, rays long and abruptly pointed; B. 5. Oxea, with a swelling at the centre

(Scale = 0.1 mm).

FIGURE 40



10. *Halina extensa* (Dendy)
(Fig. 41-43)

Stoeba extensa Dendy, 1905, p. 70, pl. 5, fig.1; Vacelet and Vasseur, 1971, p. 70, fig. 13; Halina extensa Thomas, 1986, p. 349, pl. 8, fig.14.

Material: One shell of rock oyster infested by this species from Vizhinjam (Station No. 1, Map 1).

Depth: A rock oyster shell collected from mussel bed in depths varying from 5 to 8 meters.

Colour: Pale yellow.

Description: The shell is disintegrated to the maximum and only a few openings varying in diameter from 0.5 mm to 0.8 mm could be seen at the surfaces of shell which is highly damaged. One excurrent papilla could be located; it has an apical opening (oscule) of 0.28 m (Fig. 41 C). The opening is circular with a transparent membrane extending around the opening. This transparent membrane is reinforced with triaenes and spined microxeas.

Diameter of chambers, made inside the shell by the activity of this species, may vary from 0.21 mm to 1.3 mm and as compared to those formed by various species of *Cliona*, these are quite smaller. The interior of the chambers and inter chamberal canals present an etched out appearance as in *Cliona* species, but the etchings (pits) are quite smaller, 0.016 - 0.024 mm in diameter (Fig. 42 B).

The "sponge mass" found inside the chamber resembles very much to that formed by any species of *Cliona*. The diameter of such 'sponge mass' may vary considerably as per the diameter of chamber (Fig. 42 A). The 'sponge mass' is usually spherical in shape and originate from a branch (BR in Fig. 42 A) as in *Cliona* spp. and after forming a chamber it continues to run to form another chamber. A longitudinal section of the branch (Fig. 41 D) shows that the triaenes are arranged at the periphery where the concentration of spined microxeas is quite high. Sand grains are seen incorporated into the interior of these branches. The diameter of branch may vary from 0.2 to 0.9 mm. Triaenes are usually seen in the peripheral region of the 'sponge

mass' inside the chamber, while the microxeas are distributed throughout the surface and interior with preference to the mid longitudinal surfaces and axial parts of the branch. Such an arrangement may become much diffused as it comes to the lateral surfaces of the "sponge mass" (Fig. 42 A). Branches formed from this "sponge mass" are flat (Fig. 41 A, B) and each branch, at its initial stage of growth, may put forth many lateral conical processes, but these conical processes disappear in due course as and when a new chamber is formed. Very young triaenes may be present at this stage irregularly distributed and the spined microxeas take an axial course along the new branch; sometimes even along the direction of the lateral processes (Fig. 41 A).

It could be seen that some slender branchlets are formed from the branch (Fig. 41 A, BRL) or a 'sponge mass' destined to form a chamber (Fig. 41 B, BRL). The diameter of these branchlets may vary from 0.04 to 0.37 mm and as they penetrate through the shell take a helicoidal pattern of branching (ie. branchlets are formed from one side only). Each branchlet later gets flattened and thus a new chamber is formed (Fig. 41 E). The branchlet further grows to form new chambers.

Spicules: 1. Triaenes (Fig. 43 A 1): Shaft conical; length upto 0.12 mm, clads at right angles to the shaft, may or may not divide, when they divide the branches may or may not be similar; chord length upto 0.168 mm; width of clad (at its origin) upto 0.016 mm when well developed.

2. Spined microxeas (Fig. 43 B 2): Minutely spined, length upto 0.016 mm, fairly abundant.

Remarks: *Halina plicata* which also is a boring sponge, can be differentiated well from the present species in respect of having larger triaenes and also in the presence of streptasters (in *Halina extensa*, it is microxea). *Halina extensa* is here confirmed as a boring species capable of boring into calcareous objects.

Distribution: Previously known only from the Gulf of Mannar (Dendy, 1905); it is here recorded from Vizhinjam (Arabian Sea), as a pest of rock oyster.

Fig. 41

Halina extensa (Dendy)

A. The main branch (BR) after forming a chamber runs further. From the 'sponge mass' inside the chamber, two lateral branches are given off in the opposite directions, the upper one is given in full. From the main branch (BR) a branchlet (BRL) is formed (Scale = 1 mm).

B. Initial stage in branchlet formation. This branchlet (BRL), as it goes, rebranch and form additional chambers (Scale = 1 mm).

C. Summit of oscular papilla with oscular opening (partly contracted), viewed from above. Oscular opening is reinforced with spicules (Scale = 0.1 mm).

D. Longitudinal section of a branch (older part) showing the arrangement of spicules. Sand grains are seen incorporated (Scale = 0.1 mm).

E. Branchlet (BRL), shown in Fig. B, may grow further and become stronger and may give off branchlets to form new chambers. One branchlet, thus given off, is shown (one which grows downwards) (Scale = 0.1 mm).

BR- branch; **BRL**- branchlet

FIGURE 41

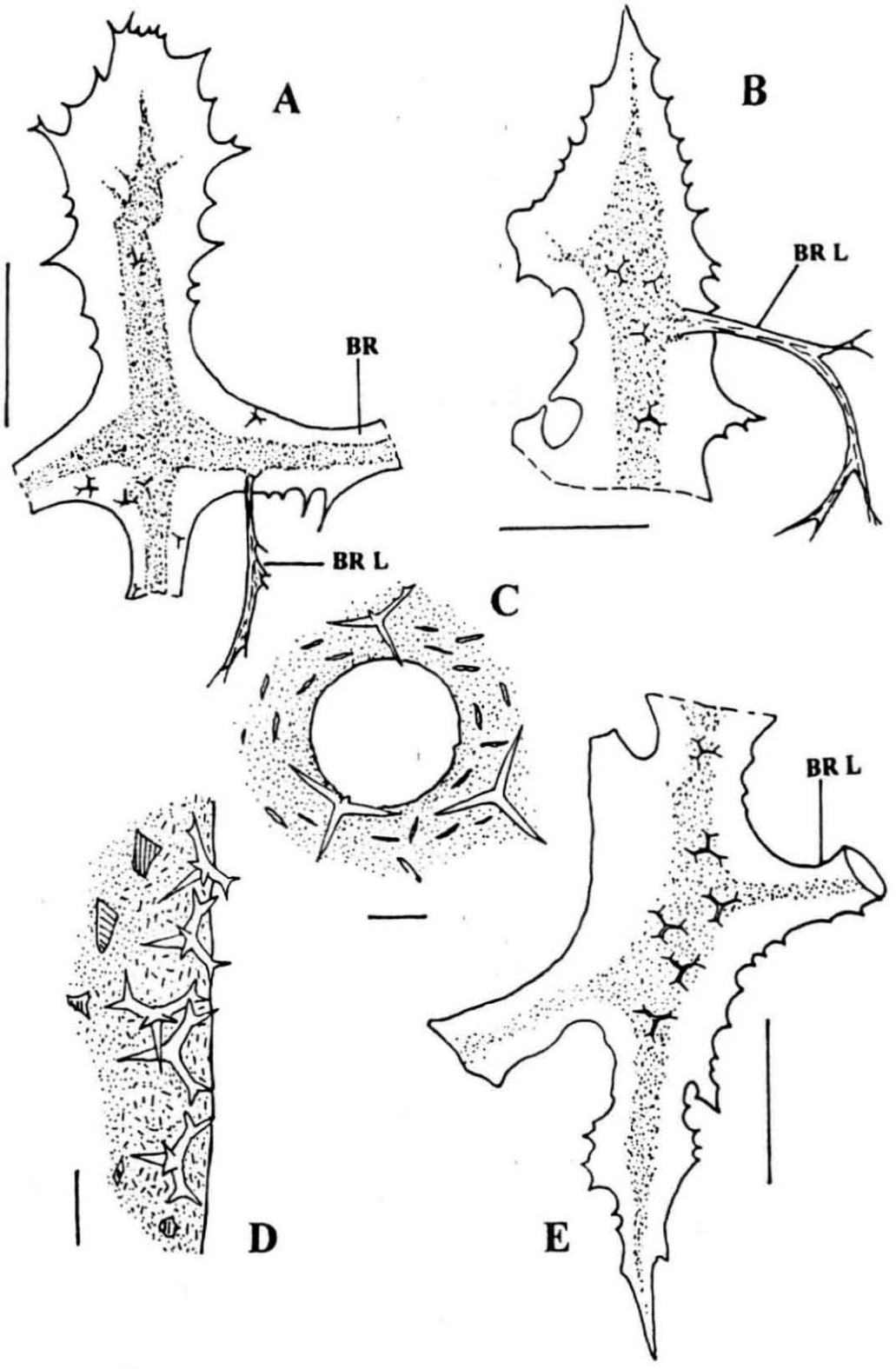


Fig. 42

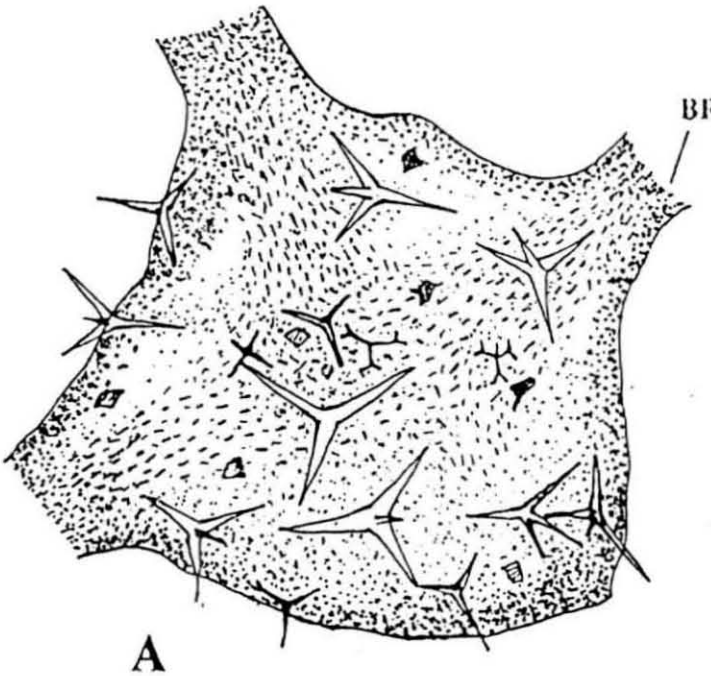
Halina extensa (Dendy)

A. Magnified view of "sponge mass" inside the chamber. The main branch (BR) after forming the "mass", continues to form the next chamber. Spined microxeas are seen through out the surface of branch with preference to mid longitudinal surfaces and axial parts. Sand grains are seen incorporated into the "sponge mass" (Scale = 0.1 mm).

B. A thin horizontal section of the shell through a chamber (CH) and two inter chamberal canals (ICC). The etched out interior clearly indicates that this species is a true boring sponge (Scale = 0.1 mm).

BR- branch; **CH**- chamber; **ICC**- interchamberal canals

FIGURE 42



A

B

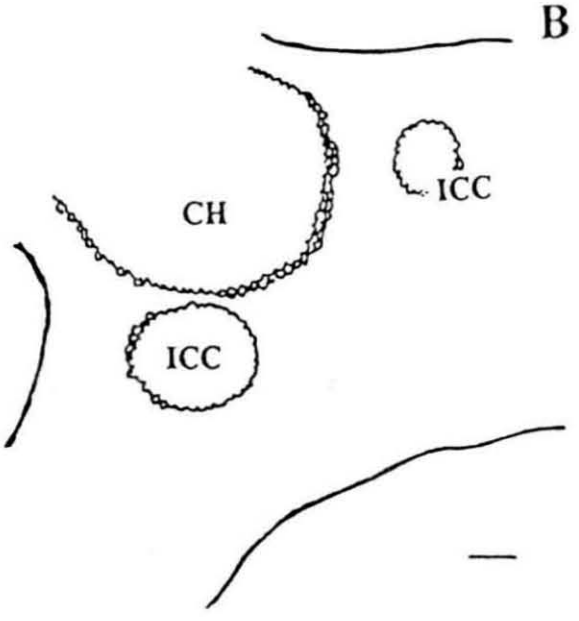


Fig. 43

Halina extensa (Dendy)

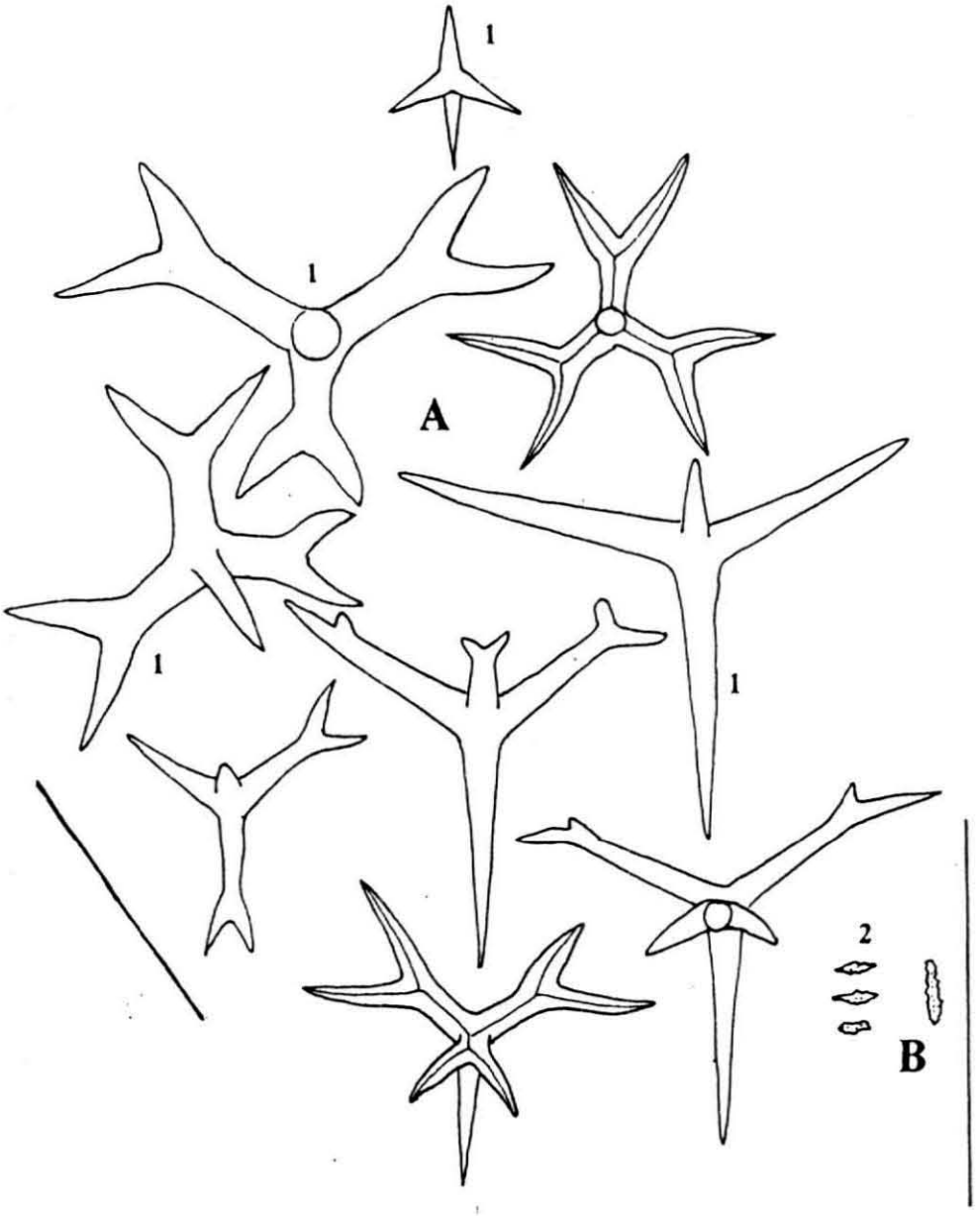
Spicules:

A 1. Trianes, different stages in the development.

B 2. Spined microxeas.

(Scale = 0.1 mm).

FIGURE 43



3. 4. Zoogeography and conclusion

In order to study the global distribution of the various species of sponges collected from the southwest coast of India as pests of mussels, their occurrence widely separated zoogeographic realms such as the Atlantic Ocean, Mediterranean Sea, Red Sea, Indian Ocean, Australian Region and the Pacific Ocean was considered. The study revealed that two species (*Cliona celata* and *C. vastifica*) are cosmopolitan in their distribution and the latter is common in the estuarine areas of all the zoogeographical realms mentioned above due to its low-salinity tolerance.

After excluding the above two species, the zoogeography of the other, seven species, may be summarized as follows.

1.Those distributed in the Atlantic Ocean
   1. *Cliona lobata*
   2. *C. carpenteri*
   3. *Thoosa armata*
   4. *Alectona millari*

2.Those distributed in the Mediterranean Sea
   1. *Thoosa hancocki*
   2. *Alectona millari*

3. Those distributed in the Red Sea
   1. *Thoosa armata*
   2. *T. hancocki*

4 Those distributed in the Australian Region
   1. *Cliona margaritifera*
   2. *C. carpenteri*

5. Those distributed in the Pacific Ocean
   1. *Cliona lobata*
   2. *C. carpenteri*
   3. *Thoosa hancocki*

Summarizing the zoogeographical affinity of the above seven species (excluding two cosmopolitan species), it may be stated that boring sponge fauna of the

southwest coast of India has the maximum affinity with that of the Atlantic Ocean and four out of seven (57.1 %) are common to these two areas. The next area which has more in common with the boring sponge fauna of the southwest coast is the Pacific Ocean, where three species (42.8 %) are common with these two areas. Affinity with the Mediterranean Sea, Red Sea and the Australian Region is the same; two species out of seven (or 28.5 %) are common with these three areas and the southwest coast. The general distribution of the various boring species in the centres selected for the present study is dealt with in Chapter IV.

4.SPECIES COMPOSITION, DISTRIBUTION AND ABUNDANCE OF BORING SPONGES

4. 1. Historical account of sponge infestation in the Indian molluscan beds

Numerous representatives of 12 major taxa of marine plants and invertebrate animals are known to excavate hard calcareous substrata by either chemical or mechanical means or by a combination of both. During the past century, a great number of papers has been written (Clapp and Kenk, 1963) dealing with the systematics, distribution, ecology and physiology of these organisms, and with the geological, chemical and biological effects they cause by contributing to coastal erosion, by influencing the calcium balance in the sea, and controlling the structure of marine communities where calcium carbonate producing organisms are dominant. An important summary of the latest research results was presented in a symposium (Carriker *et al.*, 1969) in 1969.

Collectors of marine life everywhere have been impressed by the frequent evidences of holes and caverns in a wide variety of submerged calcareous objects. The literature, beginning with the nineteenth century, contains numerous descriptions of such submerged calcareous objects, limestone, marble, coral, molluscan and barnacle shells, riddled with minute perforations and cavernous galleries containing a radially symmetrical organism. Based on collections in the Firth of Forth, Scotland, Grant in the year 1826 described the organism as a new zoophyte and named it *Cliona celata* Grant. By 1849, Hancock showed that this organism is definitely a sponge and continued the work of describing many new species of boring sponges. Hancock failed, as well as others since him, to explain how the sponge functions as a borer of calcareous material. Gray, in 1867, organised all of the boring sponges into one family, the Clionidae, but later workers, probably through uncertainty, occasionally inserted non-boring sponge species. In 1936, de Laubenfels pointed out that the family Clionidae contains only the boring sponges, "which actually perforate calcareous objects in order to establish caverns or galleries to live in. The similarity of certain non-boring sponge in skeleton characteristics or spiculation to boring sponge is not a valid reason for inclusion in the

family (Old, 1941)".

In recent years, oystermen, conservation officials and marine investigators have come to recognize the importance of boring sponges mainly through their abundance and their probable connection with the widespread depletion of certain formerly prolific oyster beds, such as that experienced in the Little Choptank region of the Chesapeake Bay in 1934. Frequently oysters taken by tongers and dredgers have very brittle or broken shells, and the oyster beds themselves contain high percentages of riddled empty shells. Typically, these broken infested shells yield conspicuous yellow to orange excavations. Because of this situation many complaints have arisen over a wide oyster producing area, all centering on sponge infestations (Old, 1941).

A few species of *Cliona* were reported from India prior to 1900. Hornell (1904) in his report on Ceylon Pearl Oyster Fisheries recorded sponges "as an enemy of pearl oyster in Ceylon pearl banks since they cause damage in two ways: first, by causing thickened deposits of nacre and other irregularities, and hence disturbance of function at the attachment of the great adductor muscle, and secondly as honey combing the shell in all directions, rendering it so rotten that it can no longer hold together." According to him (Hornell, 1904), "it is a disease of adult life, for young shells never harbour *Cliona*." Hornell also found that "whenever the inroads of *Cliona* were extensive the sub epidermal tissue in particular, and the other tissues in general, were thin and diseased looking." The entire collection made by Herdman from the pearl banks of Ceylon (= Sri Lanka) in 1902 were studied by Dendy (1905) who recorded only one species, *Cliona margaritifera* Dendy, new to science with an infestation rate as high as 80%. Dendy (1905) further reported that "*Cliona margaritifera* spread in Pearl Banks almost like an epidemic and destroyed the pearl oyster beds either partly or completely."

Since the publication of Dendy's monograph in 1905, several publications dealing with Demospongiae in general and Clionidae in particular from the Indian Seas have appeared. In this context mention may be made to the detailed work of Annandale (1915 A) dealing with "The Indian boring sponges of the family Clionidae. " This is an outstanding contribution in this field not only for the notable additions to the fauna

but also for the elaborate key and comprehensive account of 18 species of *Cliona* from the Indian Ocean. Our knowledge in this line is further elaborated by the subsequent work of Annandale (1915 B) on "Some sponges parasitic on Clionidae with further notes on that family".

With a view to studying the boring sponges affecting the coral reefs and commercially important molluscs, Thomas (1972, 1979) took up two studies subsequently. Of these the first Thomas (1972) which deals with the boring sponges from the fringing reefs of the Gulf of Mannar and the Palk Bay, recording 20 species under four orders, four families and 10 genera. Out of 20 species described in this paper one was new to science and six were new records to Palk Bay and five to the Gulf of Mannar. Besides detailed description of various species, an elaborate key to their identification is also provided in this paper. Subsequently the second paper (Thomas, 1979 B) dealing with "Boring sponges destructive to economically important molluscan beds and coral reefs in the Indian seas" appeared. Details of 32 species with salient illustrations were provided in this paper. Besides the bored shells collected from the Gulf of Mannar and the Palk Bay, shells obtained from the Andaman and Nicobar Islands, Nagapatnam, the Gulf of Kutch, Mangalore and Sri Lanka were also utilized in this study. The activity/ out burst of various species occurring in the Indian seas, boring pattern, damage caused to the shell, control measures, etc. were also discussed at length in this paper.

In order to study the distribution of boring sponges in the estuaries, their incidence pattern, species in relation to those in the adjacent marine environment, two studies were taken up by the same author on sponges from Goa. Of these, the first one is on "Boring sponges in the Zuari and Mandovi estuaries of Goa"(Thomas, 1975), and the other deals with the "An ancient windowpane oyster bed in Goa with comparative notes on the oysters in an extant bed (Thomas and Thanapathi, 1980)." Both these studies revealed that in the Indian estuaries *Cliona vastifica* Hancock is the only pest of the mollusc as in many parts of the world due to its euryhaline nature. In the marine environment of Goa, four species of boring sponges were found to infest the

oyster *Crassostrea cucullata* with the rate of infestation as high as 80 %. *Cliona celata* could succeed in colonizing the *Crassostrea* population in the estuaries of Goa during summer months when the salinity as high was as that of the adjacent sea. But in the monsoon period *Cliona celata* gets completely depleted from these estuaries as the salinity falls down to about 5 ppt, which is well within the tolerance limit of *Cliona vastifica*. Thomas and Thanapathi (1980) further showed that in the estuarine regions of Goa *Cliona vastifica* dominated in the past (in the 16 th century) as at present. Various groups of boring animals infecting the gregarious as well as the tended stocks of molluscs (sacred chank, mussels, pearl oyster etc.) of the southeast and southwest coast of India were investigated subsequently by Thomas *et al.*, (1983). This study revealed that four groups of animals (viz. sponges, molluscs, polychaetes and sipunculids) usually infest the shells of various gregarious molluscs mentioned above. Of these four groups, sponges were found rather widespread, causing considerable damage to the molluscs both in the wild and the tended stocks. A total of six species of boring sponges were reported in this paper (Thomas *et al.*, 1983). Two species of boring sponges (*Cliona lobata* and *C. margaritifera*) could be collected from Vizhinjam mussel and pearl oyster culture rafts after a long interval from their first occurrence in the Indian seas. *C. margaritifera*, after its first appearance creating an epidemic in the Sri Lankan Pearl Oyster beds in 1902 (Dendy, 1905), disappeared totally from the Indian molluscan beds. The reappearance of this highly destructive species on raft-cultured molluscs at Vizhinjam, after a long lapse of about 80 years, is really a cause of much concern, as it is not sure whether the destructive phase as has been noted in Ceylon pearl oyster beds in 1902, would ensue or not. Similarly the other species, *Cliona lobata*, is a wide-spread oyster pest of the Atlantic. Though it is recorded from the Gulf of Mannar by Burton, in 1937, no subsequent workers could record the presence of this species from the Indian seas. The reappearance of this species on raft-cultured molluscs at Vizhinjam in large numbers, hence, poses a serious threat to the mussel and oyster population in the Indian Seas as a whole.

The incidence (infestation/ 100 shells) of boring sponges noted in the natural molluscan beds was very low, say 8.5 % in the Gulf of Mannar and 4.48 % at

Tuticorin for 1931-36 period; 11 % at Thanjavur for 1952-56 period;, 5.8 % in Ramanathapuram district for 1954-58 period, 15.67 % in Shivaganga beds for 1954-57 period; 9.12 % in Kanyakumari beds for 1957-63 period and 3-8 % off Vizhinjam for 1980-82, but the incidence of boring sponges on the culture rafts at Vizhinjam was found to be as high as 80 % in 1981. This high incidence of boring sponges on rafts was due to the spreading of the above two new infiltrants (*Cliona margaritifera* and *Cliona lobata*) and these they have, in many ways, tried to suppress the abundance of other boring sponges (conventional species) already existing on the rafts. From raft-cultured pearl oysters and mussels, these two species started migrating to other natural beds of molluscs (of different species) in and around Vizhinjam first, and then to distant molluscan natural beds. There are grounds to believe that *Cliona lobata* migrated to Tiruchendur chank beds (southeast coast) and *Cliona margaritifera* to Tuticorin (southeast coast) pearl oyster culture rafts by 1982.

Inorder to study the spreading pattern and interaction of these two new *invaders* (*C. margaritifera* and *C. lobata*) with the conventional sponge species occurring both in the natural and culture systems along the southwest and southeast coast of India, a study was taken up to 1986 as continuation of the 1980-1982 survey (Thomas *et al.*, 1993). Molluscs such as mussel (both green and brown) from culture rafts and natural beds, rock oysters, *Chama* spp., *Spondylus* spp., *Xancus pyrum* (sacred chank) from both southeast and southwest coast, *Thias* spp. which usually occur gregariously in nature, were utilised for the above study. The two new invaders (*C. margaritifera* and *C. lobata*) became so common in the brown mussel beds in and around Vizhinjam (Mulloor and Kovalam) and their infestation was always higher when compared to that of the conventional species (*C. celata* and *C. vastifica*) occurring in the respective beds. At Kovalam brown mussel beds, the composition of *C. margaritifera* and *C. lobata* together accounted for 53.3 in 1983 while the only conventional species occurring in the bed, *C. vastifica*, recorded a composition of only 46.7 %. *Crassostrea* population distributed along the intertidal region of Vizhinjam recorded the occurrence of two new invaders (*C. margaritifera* and *C. lobata*) by 1983 and in 1986 about 70 % of the total number infested was by these two species, the rest of the boring species

being conventional species, *C. celata* and *C. vastifica* and these two together accounting for 30 % of the total infestation (Thomas *et al.*, 1993). *Psuedochama* shells off Vizhinjam recorded only two boring species, *C. margaritifera*, and *C. vastifica*, but *C. margaritifera* (the new invader), was found dominating (60 %) over *C. vastifica* till 1986, but later in 1987, both attained equal dominance. Shells of *Spondylus* sp. collected off Vizhinjam showed a dominance of *C. margaritifera* (the new invader) in 1986 (50 %). The other two conventional species (*C. vastifica* and *C. celata*) together accounted for the rest (50 %) of infestation. *Thais rudolphi* shells off Vizhinjam recorded the presence of the two new invaders (*C. margaritifera* and *C. lobata*) in 1982 when about 81 % of infestation was accounted by these two species. The conventional species (*C. vastifica*) was found to share about 14.3 % infestation. Another species, *Aka minuta*, was found to infest the *Thais rudolphi* population in 1982 with a percentage of 4.7 % and this was first recorded here as a boring sponge of molluscan shell (normally *A. minuta* is seen only on corals). *Thais intermedia* shells collected off Vizhinjam in 1982 did not contain *C. margaritifera* and *C. lobata* but these two species started appearing in 1986, with 60 % infestation, followed by the conventional species, *C. vastifica* with 40 % infestation. Sacred chank (*Xancus pyrum*) from the southeast and southwest coasts were studied and in all the centres only conventional species of boring sponges (*C. celata*, *C. vastifica* and *C. carpenteri*) were noticed till 1981. But by 1983 the above species composition started changing with the occurrence of *C. lobata* at Tiruchendur (southeast coast) accounting for about 20 % of the total infestation. Almost at the same period *C. lobata* could be recorded from the chank population off Quilon (Kollam) area also (Thomas, *et al.*, 1993).

The pearl oyster (*Pinctada fucata*) culture rafts at Tuticorin were infested only with two conventional species, *C. vastifica* and *C. celata* till 1981. The former dominated (88 %), followed by *C. celata* (12 %). But by 1982, *C. celata* was found to be the dominant species (83 % of infestation) making *C. vastifica* a minor constituent (15 %) among boring species. During the year 1982 the infestation of *C. margaritifera* (new invader) was detected in stray numbers (2 %). In 1987 also *C. margaritifera* could be collected from Tuticorin pearl culture rafts (Dharmaraj *et al.*, 1987). The

reappearance of *C. margaritifera* in the Gulf of Mannar may be taken as a significant event since this sponge is capable of destroying the pearl banks as had happened in 1902 at Ceylon (Sri Lankan pearl banks). Similarly the occurrence of *C. lobata*, another new invader of the Indian molluscan beds, at Tiruchendur in 1982 may also pose serious threat to the entire molluscan population of the Indian Seas, particularly of the Gulf of Mannar. Hence Thomas *et al.*, (1993) suggested a continuous monitoring on these two species (*C. margaritifera* and *C. lobata*), their spreading pattern and abundance, both in time and space, on a long-term basis.

Raft-cultured pearl oyster (*Pinctada fucata*) and flat oyster at Vizhinjam indicated an incidence (infestation/ 100 shells) of 3.8 % during 1977-1979 period (Appukuttan, 1987). However an abrupt increase in the incidence (47 %) could be noticed by 1980 and then onwards showed only an increasing trend; 60.5% by 1981, 80 % by 1982. After 1982, however, it showed a decreasing trend, 48 % by 1983 but again it went upto 57.7 % by 1984; the average incidence for the period 1980-1984 being 58 % (Thomas *et al.*, 1993).

When two new invaders, *C. margaritifera* and *C. lobata*, first appeared at Vizhinjam on raft-cultured pearl oysters in 1980, there were only two conventional species of boring sponges infecting the rafts (*C. vastifica* and *C. carpenteri*). During 1981 another conventional species (*C. celata*) made its presence felt on the raft cultured pearl oysters at Vizhinjam, and during this year the total number of boring sponge species occurring on rafts came to a total of five. While considering the contribution of each species in the total infestation, it could be noticed that the new invaders (*C. lobata* and *C. margaritifera*) contributed 70 % of the total in 1980, 66.6 % in 1981 and 66.6 % in 1982 while the other three conventional species (*C. vastifica*, *C. carpenteri* and *C. celata*) together accounted respectively for the rest (30 % in 1980, 33.4 % in 1981 and 33.4 % in 1982). The higher incidence (infestation in 100 specimen) of boring sponges noticed in 1980, 1981 and 1982 (47, 60 and 80 respectively) on the pearl culture rafts at Vizhinjam, hence may be attributed to the invasion of the two new invaders (*C. margaritifera* and *C. lobata*).

Brown mussels examined off Kovalam (natural bed) recorded 48 % incidence in 1981, 36.6 % in 1983, 10 % in 1985 and 14.7 % in 1986. The higher incidence (48 %) noticed in 1981 was due to the spreading of the new invaders into the natural populations of molluscs in and around Vizhinjam, and during this year the composition of these two new invaders accounted for a total of 66 % with *C. lobata* taking the lead (50.2 %). The other conventional species (*C. celata* and *C. vastifica*) during this year accounted for 16.6 % each and as compared with the two new invaders, these two species ie, *C. celata and C. vastifica*, showed lower incidence among the four boring sponges encountered in the bed. By 1983, *C. margaritifera* attained dominance (33.3 %) making the composition of *C. lobata* low (ie. 20 %) and this indicates that in the competition between *C. margaritifera* and *C. lobata* it is the former, that takes the advantage. *C. celata* with a composition of 16.6 % in 1981 totally disappeared from the bed and this indirectly helped *C. vastifica* to take the dominance (46.7 %) indicating that in the competition between conventional species it is *C. vastifica* which takes the lead. In 1985, *C. margaritifera* among new invaders increased its activity further and a composition as high as 45.7 % could be noticed and this, no doubt, cut down the activity of *C. lobata* to a minimum of 10 %. *C. celata* was not present in the bed and its absence probably helped to maintain the composition of *C. vastifica* at 42.5 %. Here again the dominance of *C. vastifica* over *C. lobata* is well affirmed. During 1986 the species composition in the bed changed drastically. *C. margaritifera* totally disappeared and this indirectly helped *C. lobata* to contribute as high as 50 %. *C. celata* which was not seen in the bed during 1983 and 1985 reappeared in 1986 with a composition of 21.4 % indirectly checking the activity of *C. vastifica* to a lower level of 28.6 % (see Table 3) indicating the acute competition between two conventional species *C. vastifica* and *C. celata* (Thomas *et al.*,1993). It could be noticed that there is also some competition between the conventional boring species and the two new invaders. Apart from such competition between these two groups, there is competition among various species within each group. In this "between group competition" it is new invaders which win initially but in the competition within the invader group (*C. margaritifera* and *C. lobata*) it is *C. margaritifera* that succeed finally. But in

Table 3. Year wise species composition and incidence of boring sponge at Vizhinjam during 1981-1986

Species composition	1981	1983	1985	1986
Cliona margaritifera	16.6	23.3	45.7	0
Cliona lobata	50.2	20.0	10.0	50.0
Cliona celata	16.6	0	0	21.4
Cliona vastifica	16.6	46.7	42.5	28.6
Incidence	48.0	36.0	10.0	14.7

the competition among the conventional species the one which has traditional dominance in the particular bed generally wins. When one species is inhibited directly by another it might activate a third species to multiply disproportionately causing epidemics at times. Since such an interaction is possible both in the natural and cultivated stocks of molluscs a continuous monitoring of the activities of the different boring species is essential to develop any long-term strategy (Thomas, *et al.*, 1993).

After 1986 there were no direct observation on boring sponges infesting the mussel beds off Vizhinjam or other parts along the southwest coast of India till Anitha Nair took up a study entitled " Boring sponges destructive to brown mussel population" for her M. Sc programme of M. S University (M. Sc dissertation, M. S University, Nagercoil) in 1998. For this she collected brown mussel off Vizhinjam and the study revealed an incidence as high as 82 %. Of the two new invaders only *C. lobata* could be collected by her, (and this species accounted to 39.5 %). Among the conventional species *C. vastifica* dominated (42.1 %) followed by *C. carpenteri* (13.2 %) and *C. celata* (percentage composition, 5.2 %).. The important findings emerged during her study was a total absence of *C. margaritifera* from the mussel beds off Vizhinjam and the invasion of *C. carpenteri*, which is commonly seen infesting only the cultured brown mussel at Vizhinjam.

4. 2. Sponge infestation on mussel beds during the present study period: month -wise and season -wise

1. INTRODUCTION

In order to study the infestation, distribution, abundance and species composition of boring sponges distributed along the southwest coast, mainly from Vizhinjam to Cape Comorin (Kanyakumari), six centers were selected (Map 1). The various stations were Station 1 (Vizhinjam), Station 2 (Enayam), Station 3 (Colachel),

Station 4 (Kadiyapatnam), Station 5 (Cape Comorin) and Station 6 (Mulloor).

Since the present area is under the influence of both southwest and northeast monsoons the fishing activities get suspended when the sea conditions become adverse for mussel pickers (skin divers) and restart at the onset of favourable conditions. Data, hence were collected from each center from the beginning to the end of the fishing season and thus samples for two seasons, viz. 1998 October, to 1999 April and 1999 April, to 2000 March are utilised in the present study. It is also possible that fishing activity may get suspended for some months due to adverse weather conditions. Though the period October to April is considered congenial for mussel fishing in most areas, considerable variations could be seen each year according to weather conditions. During the first season it was noticed that there was no sponge infestation on mussels collected from Station 5 (Cape Comorin) and hence the sampling for the next season was discontinued and instead another station (Mulloor) was selected as the sixth station for mussel sampling.

2. MATERIAL AND METHODS

General pattern of data collection is dealt with in the part dealing with "Material and methods". (Chapter 1). The "incidence" as given in the present account is calculated on the basis of infested shells from a total of 100 specimens collected at random and the "species composition" denotes the number of specimens infested by each species of boring sponge out of the total specimens infested. Station - wise details are given below. Both pooled data for the entire season as also monthly data are considered in the present study.

3. RESULTS

4. 2. 1. Month -wise size frequency of mussels in relation to the incidence of sponges at various centres

The incidence of boring sponge infestation is positively correlated with the size of the mussel. So the individual lengths were recorded for the samples collected from all the six stations during the period 1998-2000. Length frequency distribution helps to find the minimum size at which sponge infestation starts. The samples were grouped into class intervals of class width 5 and the length frequency distribution plotted. Mussels of size 10 mm to 14.99 mm were included in 10 mm size group and the same procedure is followed for all the individual lengths. The individual length frequencies were pooled and the length frequency was plotted to get a clear picture of the infestation pattern of the total population. The percentages of sponge-infested specimens were also plotted and the peaks are determined on the basis of numerical abundance of bored shells in various size groups.

Under this section, aspects like monthly incidence of boring sponges (infestation per 100 specimens), size -group-wise incidence of boring sponges and the size at which the sponge infestation starts etc., are considered centre -wise. Specimens, as mentioned in the section "Material and methods", were collected once or twice every month during the fishing season, which usually starts by October and ends by March/April. For example the two seasons considered at Vizhinjam (Centre-1) are as follows:

First season: October 1998 to March 1999
Second season: October 1999 to March 2000

In the following account a month-to-month comparison is made for both seasons, that is, the monthly details such as size groups of mussel fished at each

centre, size-group - wise infestation of mussel fished at each centre, size-group - wise infestation of mussel by various boring sponges, incidence for the whole month as also for each size-group of mussel etc., are compared. In other words, the above details for October 1998 (first season) are compared with those of October 1999 (second season).

Suitable bar and pie diagrams are also provided for every month to clarify the various points discussed.

Station I Vizhinjam (Figs. 44, 45)

Season I: October, 1998 to March, 1999

Season II: October, 1999 to March, 2000

October, 1998

The specimens collected from this centre varied in their length frequency distribution from 30-35 mm to 85-90 mm. Signs of sponge attack were seen in specimens falling under the size group 45-50 mm onwards with a peak in 75-80 mm size group. The incidence of infestation was 18 %.

October, 1999

During this month the specimens landed had a length frequency varying from 55-60 mm to 90-95 mm. Sponge infestation could be distinctly seen from the size groups 60-65 mm onwards with a peak in 70-75 mm size group. The incidence of infestation was 16 %.

November, 1998

The size-frequency distribution of mussel, at this centre, varied from 35-40 mm to 80-85 mm. The smallest size group containing boring sponge was at 55-60 mm, and above this size group almost all specimens were found infested by boring sponges with a peak in 70-75 mm. The incidence of infestation was 32 %.

November, 1999

Size-frequency of mussel examined during this month fluctuated between 20-25 mm and 90-95 mm. Bored shells were present in almost all size groups above 60-65 mm with a peak in 70-75 mm. The incidence of infestation was 11 %.

December, 1998

The size-group of mussel represented during the month varied from 25-30 mm to 80-85 mm. Sponge infestation could be noticed in size groups above 50-55 mm with a peak in 60-65 mm. 100 % infestation could be seen in 80-85 mm size group. The incidence of infestation was 15 %.

December, 1999

Size-groups represented in the samples, for the month, fluctuated between 35-40 mm and 100-105 mm. Sponge infested shells could be seen in 70-75 mm size group onwards with a peak in 80-85 mm. The incidence of infestation was 20 %.

January, 1999

The size-groups of mussel represented in the monthly samples fluctuated between 35-40 mm and 65-70 mm. No boring sponge could be collected from the samples during this month.

January, 2000

During this month the size groups represented in the samples varied from 40-45 mm to 85-90 mm. Bored shells could be seen in all size groups starting from 50-55 mm with a peak in 60-65 mm size group. The incidence for this month is estimated at 22.

February, 1999

The size-group of mussel represented in the samples varied from 35-40 mm to 70-75 mm. Bored specimens were not present in the samples.

February, 2000

The size-group of mussel fluctuated between 50-55 mm and 80-85 mm. Sponge infested mussels could be seen from 65-70 mm size group onwards. All specimens under the size groups 75-80 mm, 80-85 mm were infested. The incidence for the month was 6.

March, 1999

The size-groups of mussels present in the sample fluctuated between 35-40 mm and 95-100 mm. All specimens were free from the attack of boring sponges.

March, 2000

Size-group of mussel varied from 45-50 mm to 85-90 mm. Sponge infested shells were found in size groups 70-75 mm onwards. The incidence of infestation was 6 %.

Station II Enayam (Figs. 46, 47)

Season I: November, 1998 to April, 1999

Season II: November 1999 to March, 2000

November, 1998

The length frequency of mussel fished at this centre fluctuated between 50-55 mm and 110-115 mm. Infested shells were found from 65-70 mm size group

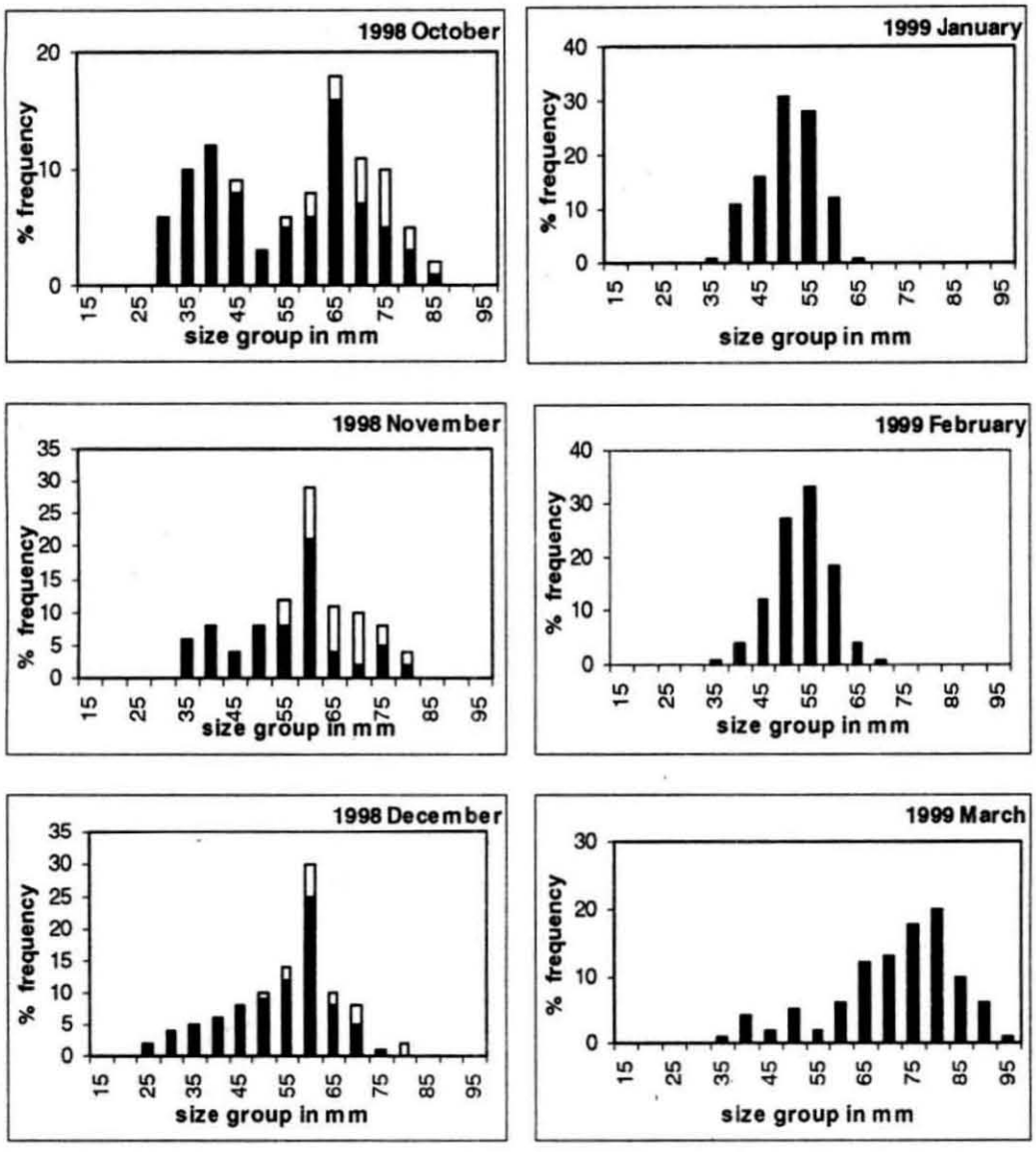


Fig. 44 Length frequency distribution of brown mussel at station I (Vizhinjam, period: October, 1998- March, 1999) ■ un-infested; □ infested

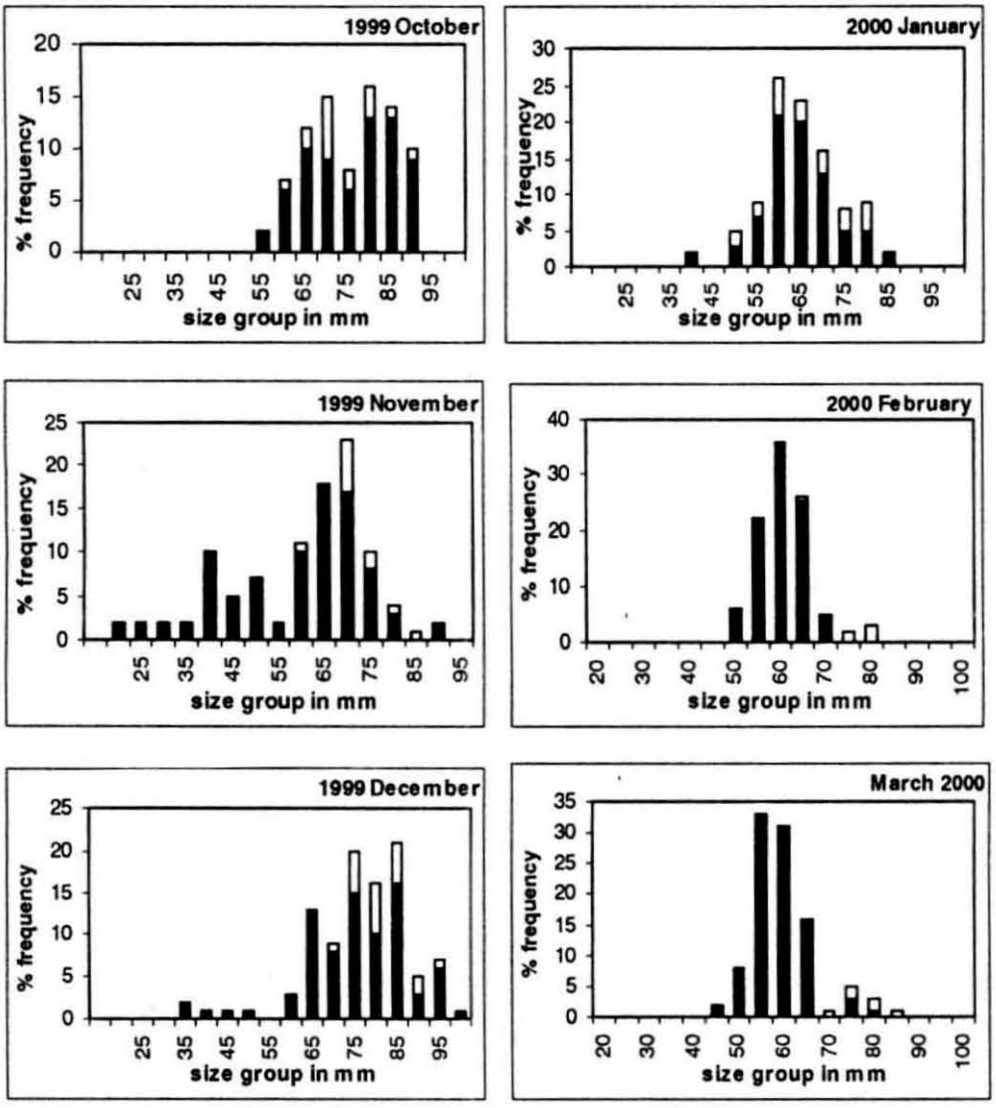


Fig. 45 Length frequency distribution of brown mussel at station I (Vizhinjam, period: October, 1999- March, 2000) ■ uninfested; □ infested

onwards up to 110-115 mm. Maximum number of bored specimens could be seen in size groups 70-75 mm and 95-100 mm. Size-group-wise incidence (%) showed an abrupt hike from 85-90 mm onwards and in 110-115 mm size group a 100 % infestation could be noticed. The incidence of infestation was 32 %.

November, 1999

The length frequency of mussel fished at this centre fluctuated between 40- 45 mm and 115-120 mm. Infested mussels were found from 80-85 mm onwards with a peak in 90-95 mm size group. Specimens falling under the size groups 110-115 mm and 115-120 mm were found infested by boring sponges rather heavily. The incidence of infestation was 5 %.

December, 1998

The length-frequency of mussel collected from this centre varied from 25 to 30 mm to 110-115 mm. Specimens measuring 65-70 mm upwards showed the presence of boring sponges with a peak in the size group 75-80 mm. All the shells falling under the size-groups 70-75 mm and 110-115 mm were infested with boring sponges. The incidence of infestation was 50 %.

December, 1999

Length frequency of mussel sampled during the month fluctuated between 35-40 mm and 100-105 mm. Infested mussels were seen only in three size groups: 65-70 mm, 70-75 mm, and 75-80 mm with a peak in 65-70 mm size group. The incidence of infestation was 6 %.

January, 1999

Length frequency of mussel sampled during the month fluctuated from 35-40 mm and 100-105 mm. Bored shells could be noticed from 60-65 mm onwards with peak in 70-75 mm size group. All the specimens in 90-95 mm and 95-100 mm size-

groups were totally infested by boring sponges. The incidence of infestation was 48 %.

January, 2000

Length frequency of mussel sampled at this centre varied from 70-75 mm to 125-130 mm. Sponge infestation was evident in all size groups above 75-80 mm and those falling above 100-105 mm were totally infested. The incidence of infestation was 85 %.

February, 1999

Length frequency of mussel, at this centre, fluctuated between 75-80 mm and 110-115 mm. Boring sponge infestation could be noticed in size groups above 80-85 mm. 100-105 mm and 105-110 mm size groups recorded 100 percent infestation. The incidence of infestation was 89 %.

February, 2000

Length frequency of mussels sampled, at this centre, fluctuated between 70-75 mm and 125-130 mm. Sponge infestation could be seen in almost all size groups with peak in 95-100 mm. All size groups above 105-110 mm were totally infested (100 % incidence) with boring sponges. The incidence of infestation was 62 %.

March, 1999

Length frequency of mussels collected at this centre fluctuated between 65-70 mm and 115-120 mm. Sponge infestation was noticed in all the above size groups with a peak in 95-100 mm. 100 % infestation was noticed only in 65-70 mm size group while in all others a percentage incidence above 75 could be seen. The incidence of infestation was 78 %.

March, 2000

Size frequency of mussels studied during this month varied from 60-65 mm to 110-115 mm. Infested shells were present in size groups 70-75 mm onwards

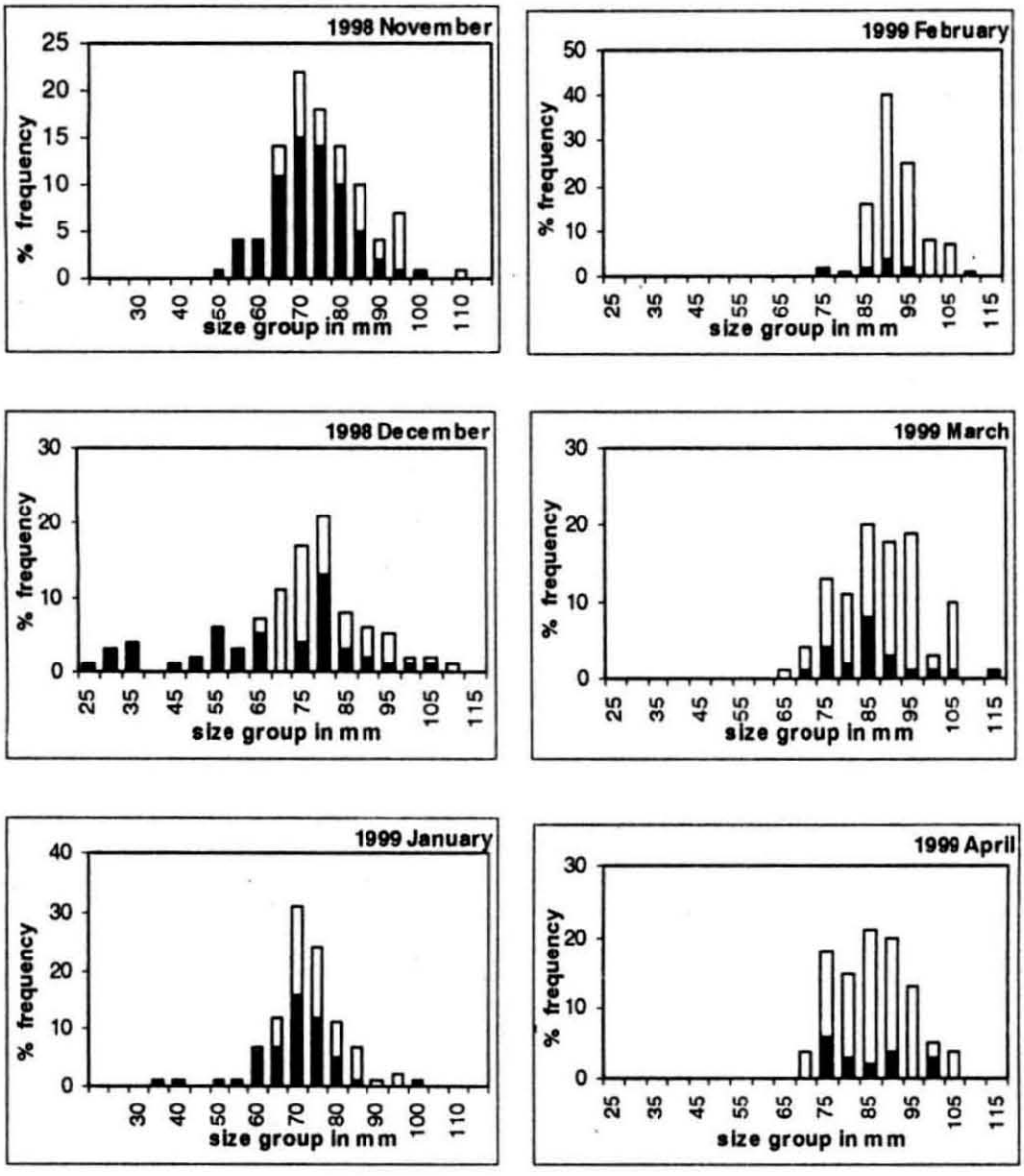


Fig. 46 Length frequency distribution of brown mussel at station II (Enayam, period: November, 1998- April, 1999) ■ uninfested □ infested

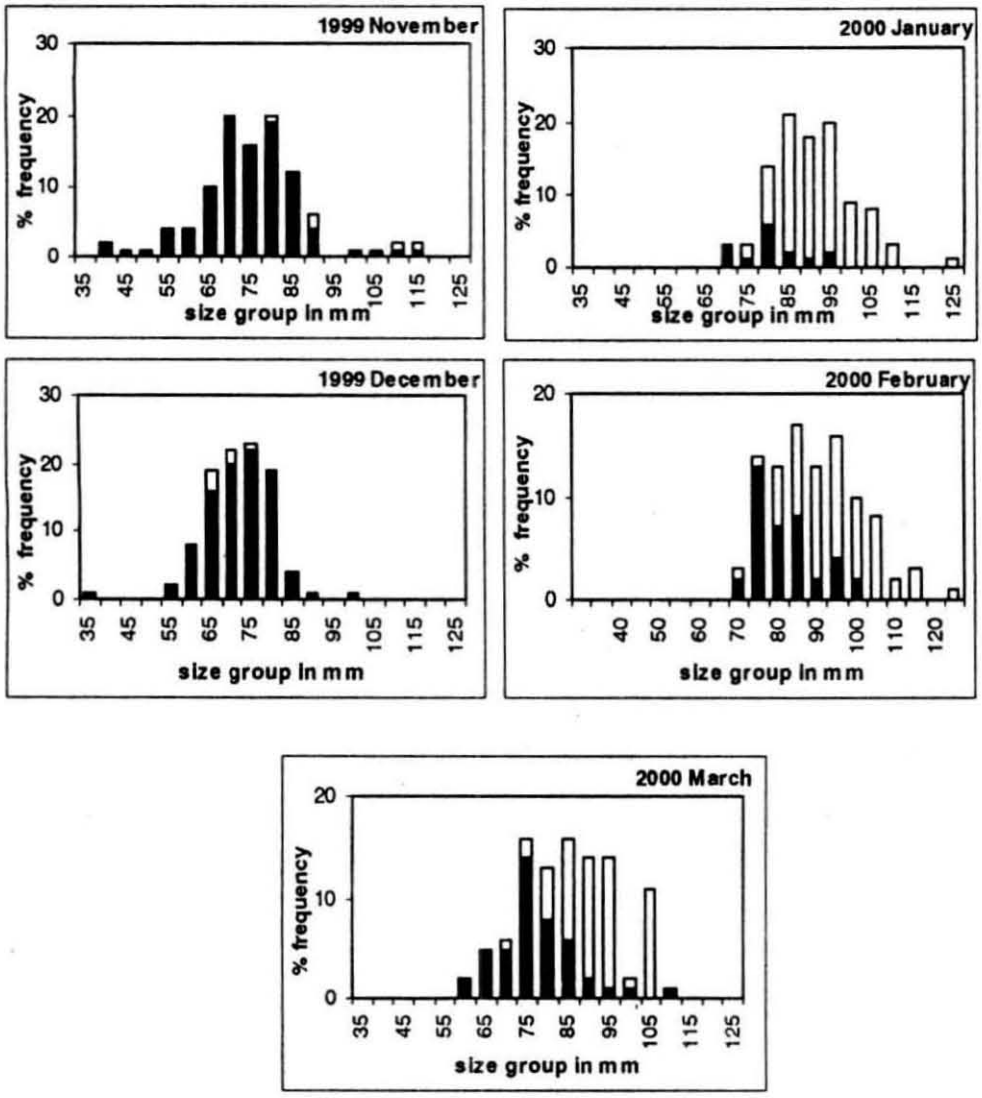


Fig. 47 Length frequency distribution of brown mussel at station II (Enayam, period: November, 1999 - March, 2000) ■ uninfested □ infested

with a peak in 95-100 mm size group. 100 % incidence was found in size group 105 - 110 mm only. The incidence of infestation was 55 %.

April, 1999

The size frequency of mussel sampled during this month varied from 70-75 mm to 105 -110 mm. Boring sponges were present in all the above size groups with a peak in 85-90 mm. 100 % infestation could be seen in the size groups, 70-75, 95-100 and 105-110 mm. The incidence of infestation was 82 %.

Station III Colachel (Figs. 48, 49)

Season I: November, 1998 to March, 1999

Season II: November 1999 to February, 2000

November, 1998

Size frequency of mussel collected from this centre varied from 40-45 mm to 80-85 mm and infested shells were present in five size groups with a peak in 55-60 mm. The incidence of infestation was only 7 %.

November, 1999

Size frequency of mussel collected from this centre varied from 60-65 mm to 85-90 mm. Infestation could be seen only in three size groups; 65-70 mm, 70-75 mm and 75-80 mm with a peak in 75-80 mm. The incidence of infestation was only 4 %.

December, 1998

Size frequency of mussel collected from this centre varied from 10-15 mm to 80-85 mm. There was no infestation in any of the shells examined during this month.

December, 1999

Size frequency of mussel collected from this centre varied from 60-65 mm to 75-80 mm. Infested shells were present in all size groups with a peak in 65-70 mm.

The incidence of infestation was 7 %.

January, 1999

Size frequency of mussel collected from this centre varied from 45-50 mm to 75-80 mm. There was no infestation at this centre during January, 1999.

January, 2000

Size frequency of specimens collected during January, 2000 fluctuated between 45-50 mm and 85-90 mm. Infested shells could be seen in size groups 70-75 mm onwards with a peak in 80-85 mm. When examined size group wise, the size groups 80-85 mm and 85-90 mm showed an incidence of 50. The incidence of infestation was 7 %.

February, 1999

Size frequency of specimens sampled at this centre varied from 65-70 mm to 115-120 mm. All size groups above 75-80 mm showed the signs of sponge infestation with a peak in 95-100 mm; 100 % infestation was seen only in 75-80 mm size group, and all the other larger size groups were infested to 80-90 % level. The incidence of infestation was 43 %.

February, 2000

The size frequency of mussel sampled during this month varied from 45-50 mm to 75-80 mm. Infested shells were seen in size groups 70-75 mm and 75-80 mm almost totally. The incidence of infestation was 4 %.

March, 1999

Size frequency of mussel examined fluctuated between 40-45 mm and 65-70 mm. No sponge infestation could be noticed during this month.

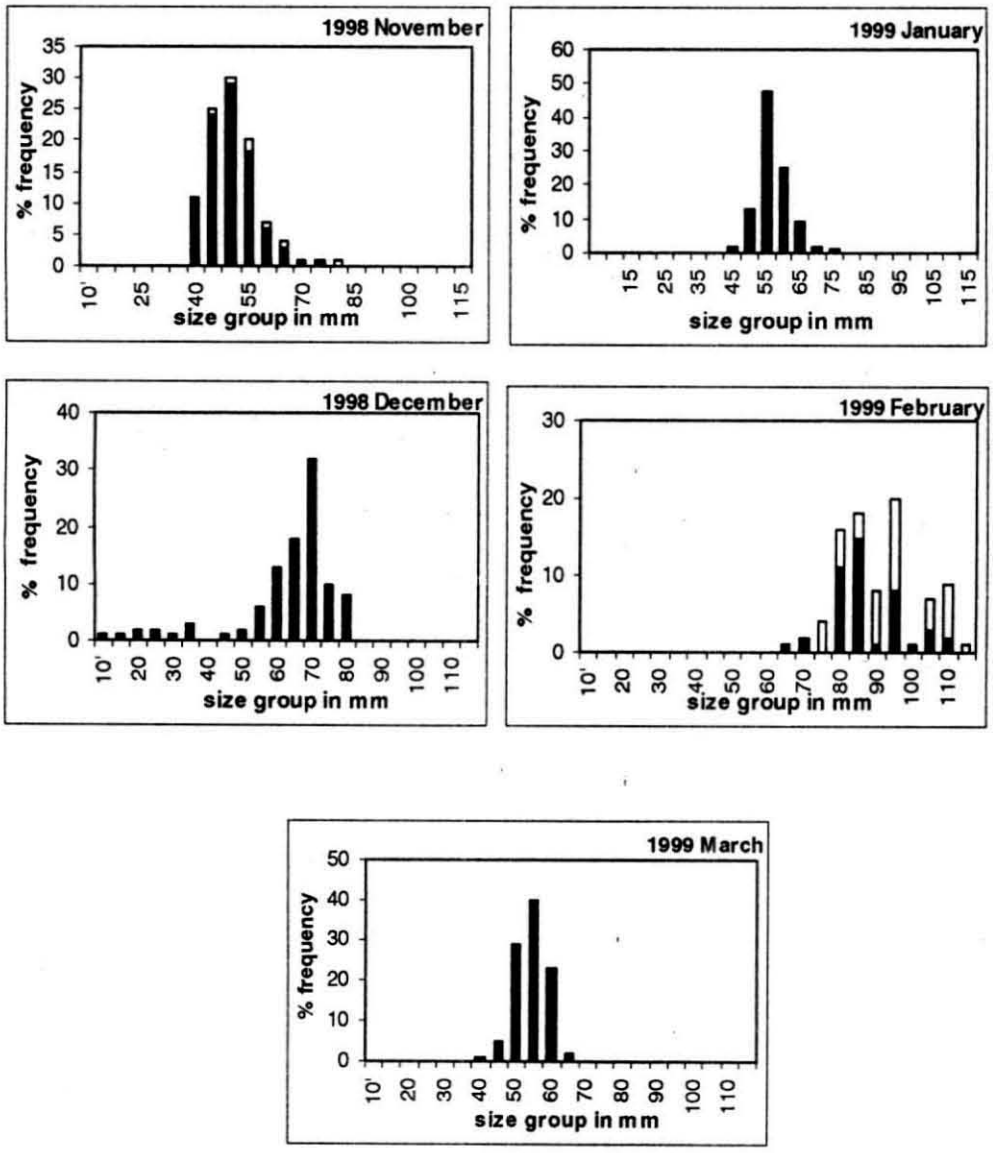


Fig. 48 Length frequency distribution of brown mussel at station III (Colachel, period: November, 1998- March, 1999) ■ uninfested; □ infested

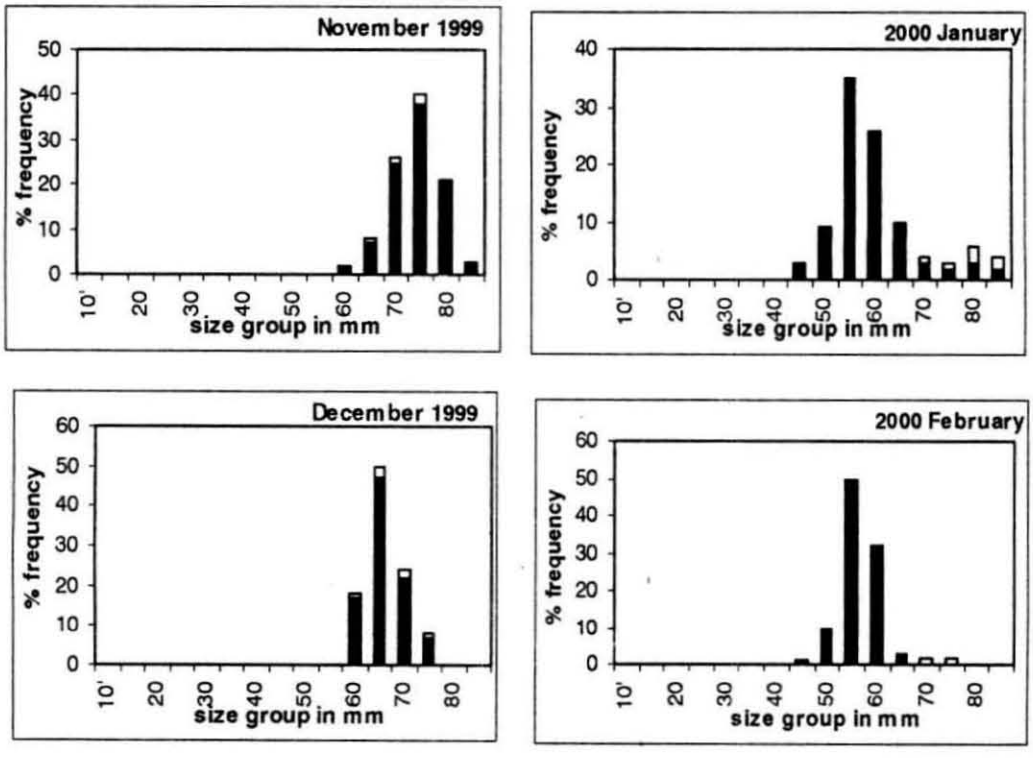


Fig. 49 Length frequency distribution of brown mussel at station III (Colachel, period: November, 1999- February, 2000) ■ uninfested; □ infested

Station IV Kadiyapatnam (Figs. 50, 51)

Season I: November, 1998 to March, 1999

Season II: November 1999 to April, 2000

November, 1998

Size frequency of specimens collected from this centre varied from 20-25 mm to 70-75 mm. No sponge infestation could be seen in the samples.

November, 1999

The size frequency of specimens collected from this centre fluctuated between 55-60 mm and 80-85 mm. Infestation could be noticed only in three size groups; 65-70 mm, 75-80 mm and 80-85 mm with a peak in 80-85 mm size group. The incidence of infestation was 4 %.

December, 1998

The size frequency of specimens collected from this centre fluctuated between 25-30 mm and 85-90 mm. Infestation could be noticed only in specimens measuring above 55-60 mm with peak in the size groups 60-65 mm and 70-75 mm. 100 % infestation was not noticed in any of the size groups mentioned above. The incidence of infestation was 11 %.

December, 1999

The size frequency of specimens at this centre varied from 60-65 mm to 90-95 mm. Infested specimens were seen in the size groups 60-65 mm to 80-85 mm and in no size group a 100 % infestation was seen. The incidence of infestation was 7 %.

January, 1999

The size frequency of mussel fished at this centre fluctuated between 40-

45 mm and 65-70 mm. Sponge infestation could not be recorded from any of the specimens examined.

January, 2000

The size frequency of specimens collected varied from 65-70 mm to 115-120 mm. Sponge infestation was present from 80-85 mm size group onwards. Infested shells could be seen in almost all size groups with a peak in 95-100 mm. 100 % incidence could be noticed only in the size group 110-115 mm. The incidence of infestation was 31 %.

February, 1999

Size frequency of specimens collected from this centre varied from 45-50 mm to 115-120 mm. Sponge infestation could be seen in shells measuring above 65-70 mm and almost all size groups above this were severely infested. Size groups 90-95 mm, 95-100 mm and 110-115 mm were infested at a 100 % level. The incidence of infestation was 48 %.

February, 2000

During this month the size frequency of mussels fluctuated between 55-60 mm and 95-100 mm. Infested shells were seen in almost all size groups with a peak in 80-85 mm.The incidence of infestation was 13 %.

March, 1999

Size frequency of mussels fluctuated between 25-30 mm and 75-80 mm. Examination of shells revealed that they are not infested by boring sponges.

March, 2000

Size frequency of specimens collected during the month varied from 55-60 mm to 100-105 mm. Sponge infestation could be seen in size groups upto 85-90 mm with a peak in 75-80 mm. The incidence for the month was estimated at 14.

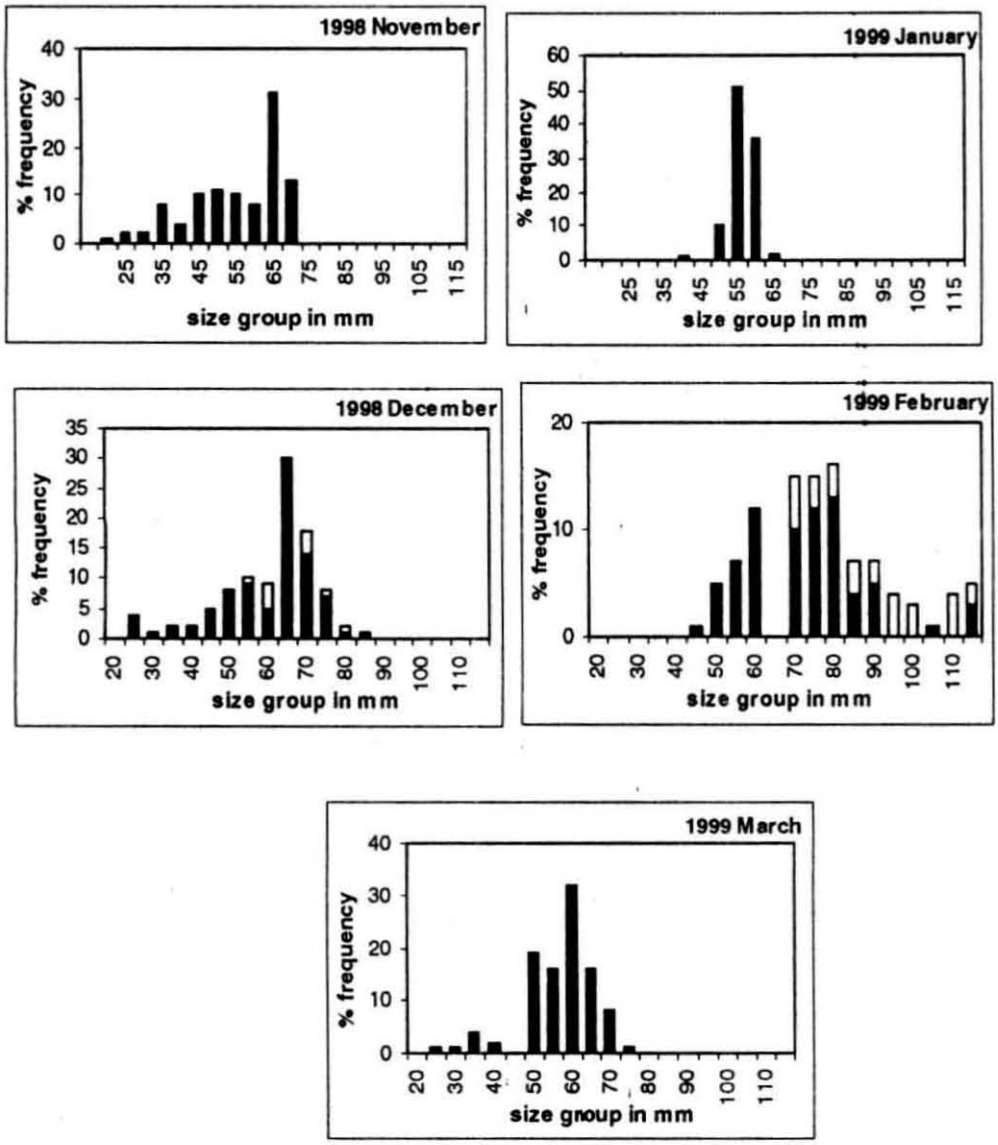


Fig. 50 Length frequency distribution of brown mussel at station IV (Kadiyapatnam, period: November, 1998- March, 1999) ■ uninfested; □ infested

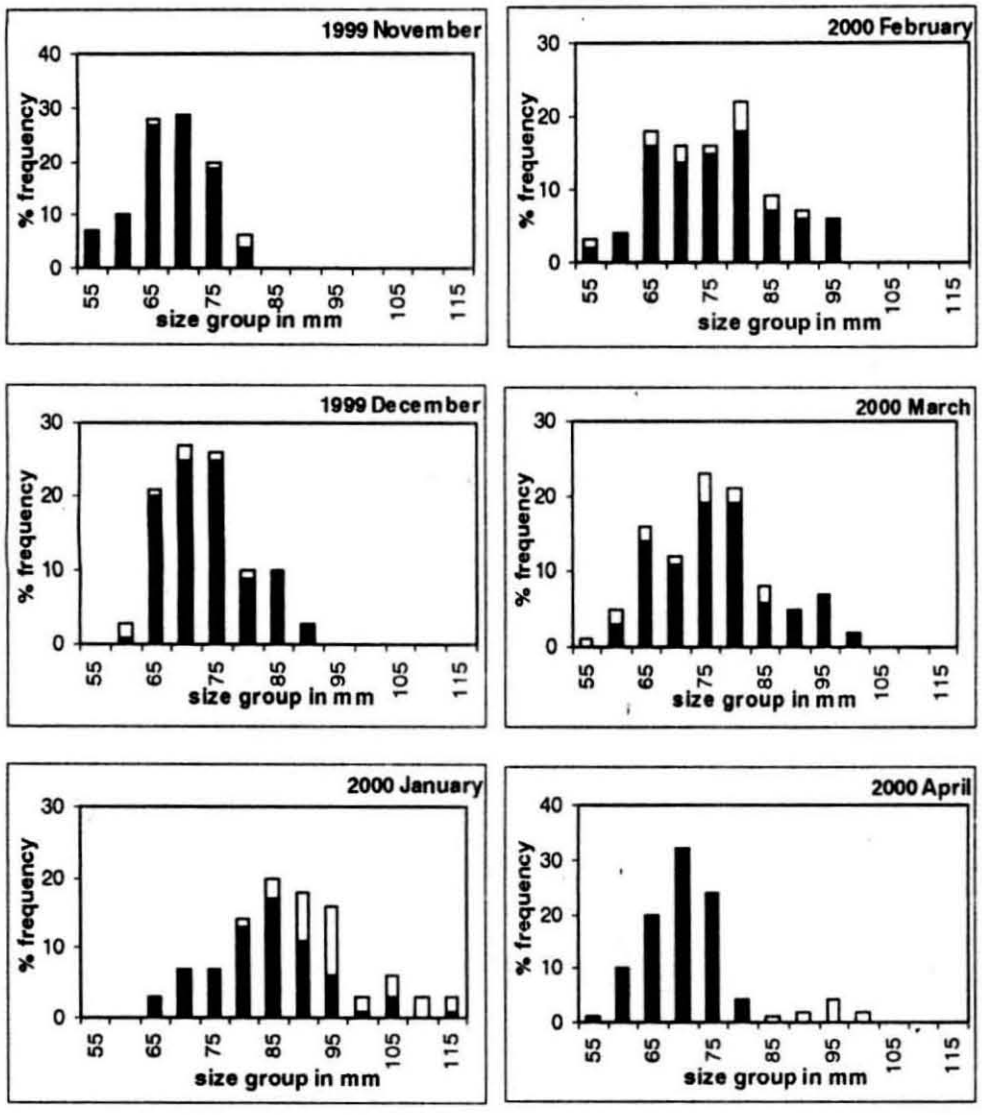


Fig. 51 Length frequency distribution of brown mussel at station IV (Kadiyapatnam, period: November, 1999- April, 2000) ■ uninfested; □ infested

April, 1999

There was no fishing at this centre during April.

April, 2000

The size frequency of mussel fluctuated between 55-60 mm and 100-105 mm. All the size groups above 85-90 mm were infested totally with a peak in the size group 95-100 mm. 100 % incidence was noticed in all size groups above 85-90 mm. The percentage incidence for this month works out to 9.

Station V Cape Comorin =Kanyakumari (Fig. 52)

Collections made during the first season indicated that the brown mussel fished at this centre are not bored by sponges and hence the sampling was discontinued at this centre during the second season.

The total length of mussel at this centre, varied from 10-80 mm and the various size groups collected are in full agreement with those collected from other centres.

Station VI Mulloor (Fig. 53)

Collections were made at this centre only from October, 1999 to March, 2000 and based on the monthly samples the details on size group wise infestation and monthly percentage incidence are presented below:

October, 1999

Size frequency of mussels collected at this centre fluctuated between 45-50 mm and 85-90 mm. Infested shells were present only in three size groups, viz. 45-50 mm, 55-60 mm and 60-65 mm. The incidence of infestation was 3 %.

November, 1999

During this month, the size frequency of mussel collected at this centre varied from 50-55 mm to 90-95 mm. Infested mussel could be seen in all size groups above 60-65 mm with a peak in 75-80 mm size group. In no size group a 100 % infestation could be seen. The incidence of infestation was 31 %.

December, 1999

Size frequency of mussels collected during this month fluctuated between 40-45 mm to 90-95 mm. Specimens measuring above 60-65 mm were only infested with boring sponges with a peak in the size group 65-70 mm. In no size group a 100 % infestation was noted. The incidence of infestation was 10 %.

January, 2000

Size frequency of specimens collected varied from 45-50 mm to 85-90 mm. Infested specimens were seen in size groups starting from 50-55 mm to 75-80 mm with a peak in 65-70 mm. 100 % infestation was not recorded in any size group. The incidence was only 10 for this month at this station.

February, 2000

Size frequency of mussel ranged from 30-35 mm to 95-100 mm. Infestation started at 65-70 mm and could be traced upto 85-90 mm with a peak in 75-80 mm. Here also 100 % infestation was not seen for any size group. The incidence of infestation at this station was only 10 for the month.

March, 2000

Size frequency of mussel, at this centre, was found to vary between 45-50 mm and 70-75 mm. Infestation started at 55-60 mm size group and was traceable upto 70-75 mm with a peak in 60-65 mm. The incidence at this station was only 5.

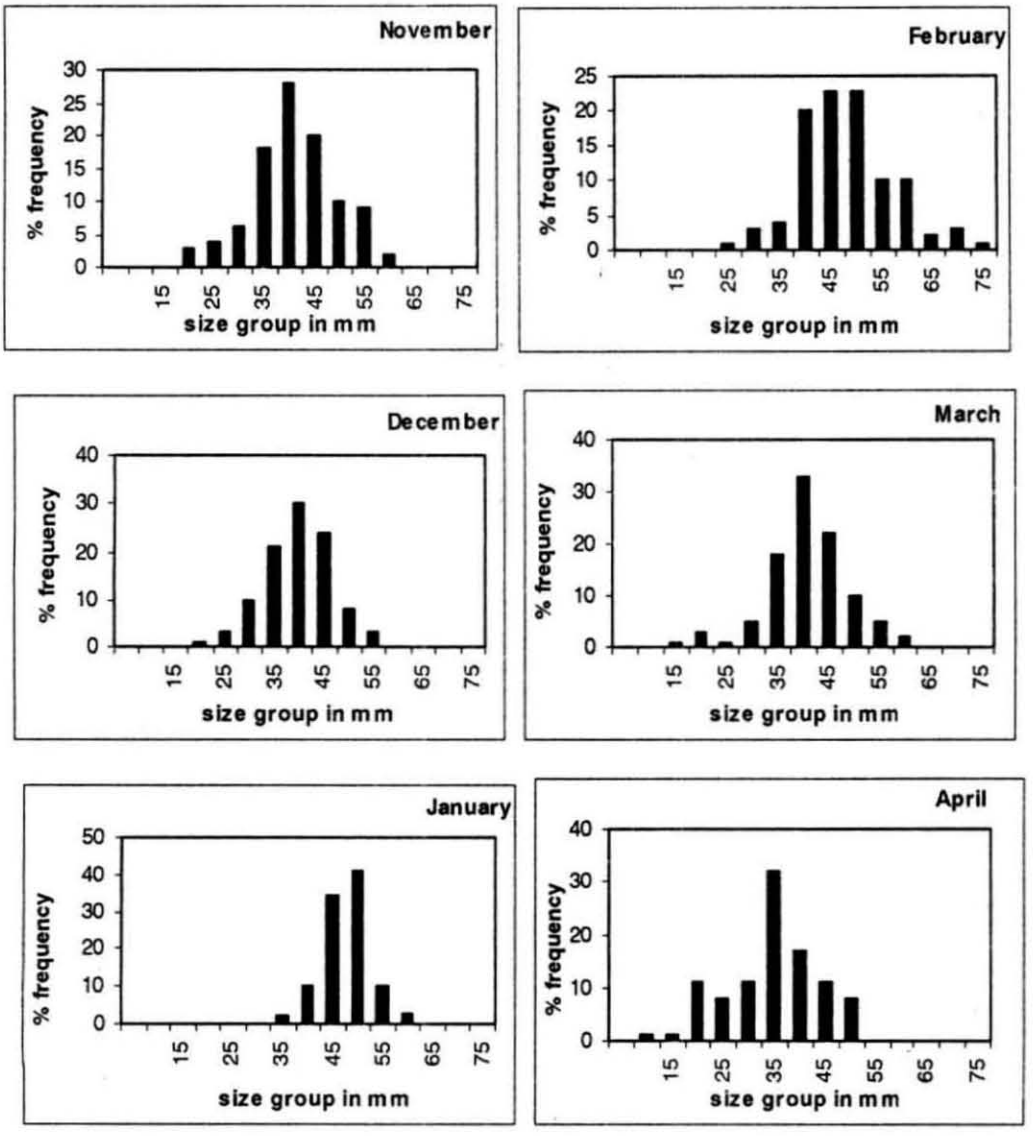


Fig. 52 Length frequency distribution of brown mussel at station V (Kanyakumari, period: November, 1998- April, 1999) ■ uninfested

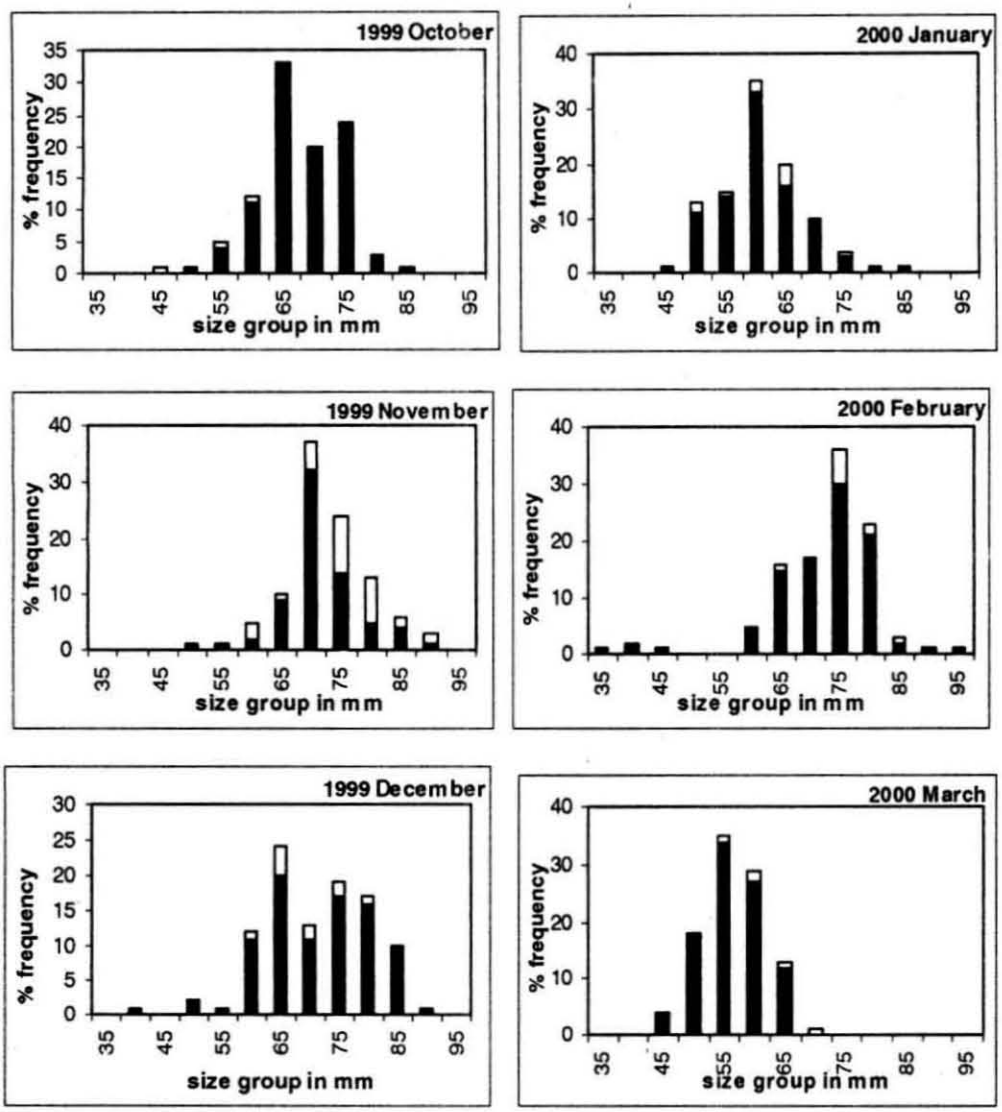


Fig. 53 Length frequency distribution of brown mussel at station VI (Mulloor, period: October, 1999- April, 2000) ■ uninfested; □ infested

4. 2. 2. Season-wise and station-wise incidence, species composition and intra specific competition of boring sponges

Station 1, Vizhinjam (Map 1)

Specimens of mussel were examined during both the seasons. It was found that the incidence was only 9.28 % during the first season, but in the subsequent season it showed an increasing trend with an incidence value of 13.16 % (Table 4, last two columns) (Map 2).

It may be interesting to note in this context that the incidence has come down to a lower level during the present study as compared to 1981 and 1982 period when the two new invaders (*Cliona margaritifera* and *Cliona lobata*) started infesting the mussel beds off Vizhinjam.

The incidence, as given in the above table, indicates that in 1981 and 1983 it was quite high, 48 % and 36.6 % respectively and obviously it was due to the spreading of the two new invaders, *C. margaritifera* and *C. lobata*. It could also be noticed that "in almost all beds the new invaders outdid the conventional species and this inturn, resulted in a sudden hike in the incidence. But this sudden spurt declined in natural beds and in artificial beds (culture rafts) the upward trend was retained for a longer period" (Thomas *et al.*, 1993). In the present study it could be seen that the incidence was only 9.28 % and 13.16 % for the first and second seasons respectively and the downward trend in incidence which started after 1985 is still in force in the mussel beds off Vizhinjam (Map 2). It may be seen from the Table (Table 4) that attack of *C. margaritifera* dwindled considerably from 1985 onwards and in 1986 it was not at all present in the molluscan beds off Vizhinjam. In the present study also the percentage composition of *C. margaritifera* was found to be quite negligible (7.9 and 6.2 respectively).

On the contrary, the other member among the two new invaders, viz. *Cliona lobata*, is represented in every year with a higher species composition often competing with *Cliona vastifica*, one of the commonest conventional species in the

Indian molluscan beds. In 1985, when the species composition of *C. lobata* decreased to 11.8 the same of *C. vastifica* increased to 42.5 % and in 1986 when the composition of *C. lobata* increased to 50 that of *C. vastifica* decreased to 28.6 % (Table 4). During the present study also a similar trend could be observed. During the first season, (October, 1998 to March, 1999) the species composition of *C. lobata* decreased to 36.5 but of *C. vastifica* increased to 50.8 %. In the next season ie. October, 1999 to March, 2000, the species composition of *C. lobata* increased to 50.6, the same of *C. vastifica* decreased to 35.8 % showing that *C. lobata*, the new invader, and *C. vastifica*, one among the conventional species, are in severe competition (Table 4).

Inorder to study the competition between and among the new invaders and the conventional species the data were analysed month wise and the same is given below in Figs. 54 A & B. During October and November (1998, the first season) both the new invaders (*C. lobata* and *C. margaritifera*) and two conventional species (*C. vastifica* and *C. celata*) were present. The conventional species *C. vastifica* accounted for the maximum in species composition in both these months with 50.7 % and 47 % respectively, followed by *C. lobata*, one of the two new invaders. *C. margaritifera* infestation was negligible with 11 % and 10 % respectively. *C. celata* infestation also was negligible (11 % and 3 % respectively). During the next month, ie. in December both *C. margaritifera* and *C. celata* were not represented. The composition of *C. lobata* increased from 27 % in October to 40 % in November while that of *Cliona vastifica* decreased from 50 % in October to 47 % in November; but this species made a substantial hike from 47 % to 60 % in December and this hike probably resulted in the disappearance of the other two species (*C. margaritifera* and *C. celata*), which were showing poor representation with decreasing trend in their species composition of previous months. During the next season (October, 1999 to March, 2000) the bed was bored only by two species in October, viz. *C. lobata* and *C. vastifica* and the latter

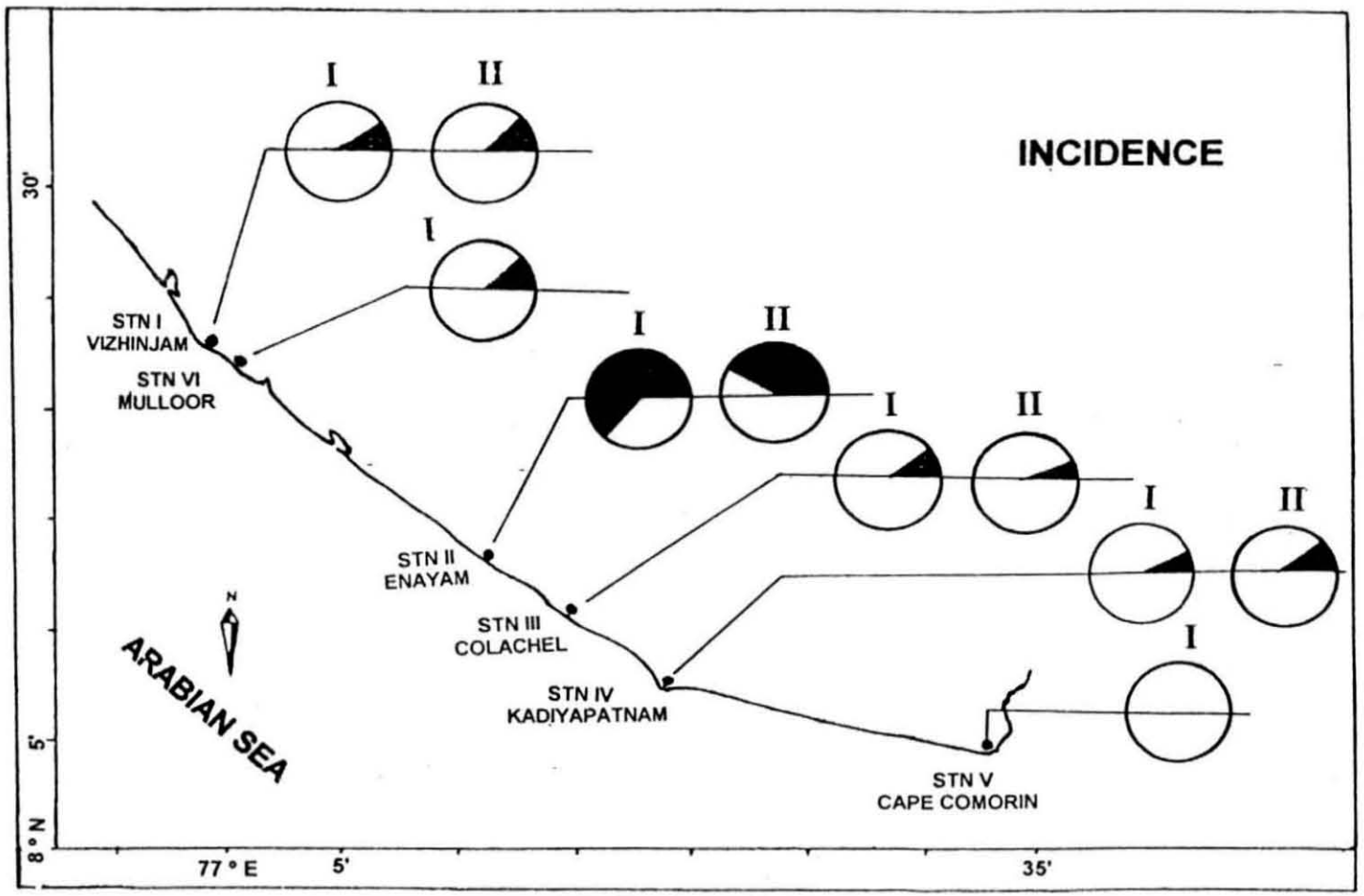
Table 4. Species composition and incidence of boring sponges at Vizhinjam natural beds, for different years and for the present period

Species composition	1981*	1983*	1985*	1986*	Ist season**	IInd season**
Cliona margaritifera	16.6	33.3	45.7	0	7.9	6. 2
Cliona lobata	50.2	20.0	11.8	50.0	36.5	50.6
Cliona celata	16.6	0	0	21.4	4.8	7.4
Cliona vastifica	16.6	46.7	42.5	28.6	50.8	35.8
Cliona carpenteri	0	0	0	0	0	0
Total %	100.0	100.0	100.0	100.0	100.0	100.0
incidence in the year	48	36.6	10.0	14.7	9.28	13.16

*Thomas *et al.*, 1993
** present study

Map 2. Incidence of boring sponges at stations I-VI

MAP 2


INCIDENCE
I
II
STN I
VIZHINJAM
STN VI
MULLOOR
STN II
ENAYAM
STN III
COLACHEL
STN IV
KADIYAPATNAM
STN V
CAPE COMORIN
ARABIAN SEA
N
30'
5'
8° N
77° E
5'
35'

formed the dominant species (69 %) in October but the composition changed by November and *C. lobata* composition increased to 90.9 % and this trend was traceable upto January, 2000. Though *C. lobata* dominated, it showed a decreasing trend from 90.9 % in November, 1999 to 59 % in January, 2000. But at the very same time *C. vastifica* showed an increasing trend from 9.09 % (species composition) in November, 1999 to 41 % in January, 2000 and to 50 % by March, 2000. Though *C. margaritifera* (new invader) and *C. celata* (conventional species) were present in the mussel beds in December, 1999 in negligible proportion (15 % and 5 % respectively) they became totally absent by January, 2000.By February, 2000, *C. lobata* disappeared totally and the species found were C. *margaritifera* (new invader) and the two conventional species, *C. celata* and *C. vastifica* with an equal share (33.3 % each). By March, 2000, *C. margaritifera* also disappeared totally and the species composition was equally shared by the two conventional species, *C. vastifica* and *C. celata* at 50 % level.

Station II, Enayam (Map 1)

It is a centre were the maximum number of boring sponge species (totally 8) could be collected (Map 3).

From this centre, *Aka minuta* is recorded as a pest of mussel for the first time and *Alectona millari*, as a record to Indian Ocean.

Specimens of mussel were examined during both seasons. It could be seen that the incidence was 63.16 % and 42.6 % respectively during the first and second seasons. As compared to the first season, the second season recorded a lower percentage incidence (Map 2). It may be stated that maximum percentage incidence was recorded at this station during the present study period.

Since no study on the boring sponges of the station was made in the past it is difficult to assess the present status of infestation at this centre. It is evident that both *C. margaritifera* and *C. lobata* have spread to this station and are competing with

the conventional species available here. Infestation of *C. margaritifera*, one of the two new invaders, was not severe and the composition of this species was 18.7 % and 10.3 % respectively during the first and second seasons (Table 5) but when compared with those at Vizhinjam (Station 1) the composition noted here is at a higher level. *C. lobata* one among the two new invaders, constituted the dominant species among the boring sponge species with a composition of 52 % in the first season while it occupied the second position in the subsequent season (Table 5) with a composition of 39.4 %. *C. vastifica*, one among the conventional species, occupied the first position during the second season with a composition of 44.6 % considerably improving its position noted during the first season (22.7 %) (ie. November, 1998 to April, 1999). All the other conventional boring species, viz. *C. celata*, *C. carpenteri* etc. were represented by very low composition. Here also, as seen at Vizhinjam (Station I), the competition between *C. lobata* (new invader) and *C. vastifica* (conventional species) was quite severe and these two have practically suppressed all the other species of boring sponges in these beds (Table 5).

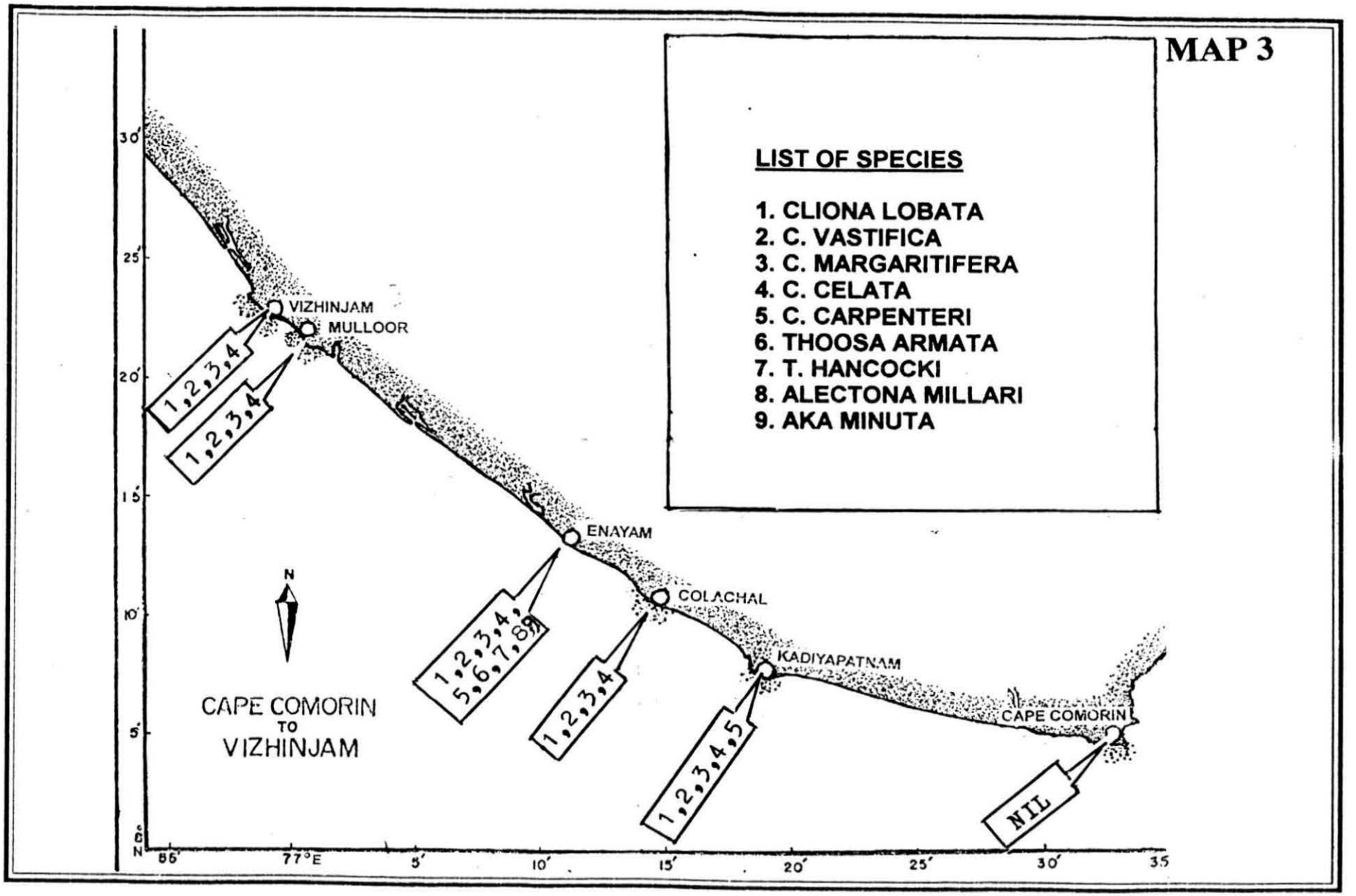
To collect more information on the competition between conventional species and the new invaders and also the competition between the species of the above two groups (conventional and new invaders) the data collected were analysed month wise and the same is given in Figs. 54 C & D. At this station, *C. lobata* (new invader) was dominating throughout the first season while in the second season, *C. vastifica* dominated partly, ie, during November, 1999 to February, 2000 and then the dominance shifted to *C. lobata* by March, 2000. *C. margaritifera* infestation noted was comparatively higher in the first season and accounted to about 31.2 % in November, 1998 and then declined to 8.5 % by April, 1999.

During the second season *C. margaritifera* was present only for three months (January-March, 2000) with species composition ranging from 3.2 % (in February) to 18.2 % (March). All the other species, at this station, recorded only negligible composition (Figs. 54 C & D). Competition of *C. lobata* with *C. vastifica* was quite evident at this station also.

Table 5. Season - wise species composition at Enayam (Station II)

Sl. No.	Species	1 st season composition (%)	11 nd season composition (%)
1	*Cliona lobata*	52.0	39.4
2	*Cliona vastifica*	22.7	44.6
3	*Cliona margaritifera*	18.7	10.3
4	*Cliona celata*	2.9	2.8
5	*Cliona carpenteri*	2.1	0.5
6	*Thoosa spp.*	1.0	0
7	*Alectona millari*	0.5	0
8	*Aka minuta*	0	2.3

Map 3. Map showing the list of species identified from Stations I-VI


MAP 3
LIST OF SPECIES
1. CLIONA LOBATA
2. C. VASTIFICA
3. C. MARGARITIFERA
4. C. CELATA
5. C. CARPENTERI
6. THOOSA ARMATA
7. T. HANCOCKI
8. ALECTONA MILLARI
9. AKA MINUTA
VIZHINJAM
MULLOOR
ENAYAM
COLACHAL
KADIYAPATNAM
CAPE COMORIN
CAPE COMORIN TO VIZHINJAM
1,2,3,4
1,2,3,4
1,2,3,4,5,6,7,8,9
1,2,3,4
1,2,3,4,5
NIL
N
8° 30'
25'
20'
15'
10'
5'
8° 00' N
77°E
5'
10'
15'
20'
25'
30'
35

Station III, Colachel (Map 1)

Colachel is a centre where only four boring species could be collected (Map 3); two new invaders (*C. margaritifera* and *C. lobata*) and two conventional species. Mussel shells were examined during both seasons (first season; November, 1998 to March, 1999); (second season; November, 1999 to February, 2000) and the incidence recorded was 10 % during the first season and 5.5 % during the next season (Map 2). Though the two new invaders (*C. margaritifera* and *C. lobata*) made their presence at this centre, no observable hike in the incidence could be noticed. Since there are no earlier studies on the sponge infestation on brown mussel from this centre, it is difficult to compare and contrast the infestation seen at present with reference to any in the past.

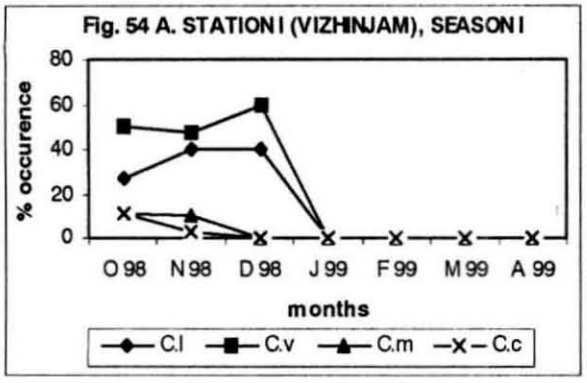
An examination of the pooled data collected from this centre for both seasons indicates that the conventional boring species of the Indian molluscan beds, *C. vastifica*, dominated during the first and second seasons with a composition of 44 % and 45.5 % respectively (Table 6). Among the two new invaders only *C. lobata* was conspicuous with a species composition of 40 % and 31.8 % respectively during the first and second seasons and this species was closely in competition with *C. vastifica*, the conventional species. *C. margaritifera* registered very low % compositon with 8 % in the first season and 4.5 % in the second season.

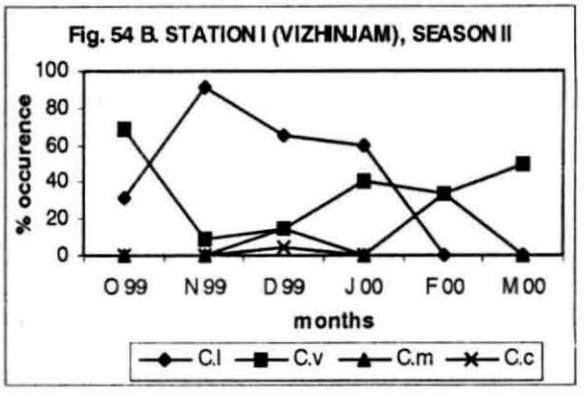
The other conventional species, *C. celata*, though poorly represented in the first season (composition 8 %), showed some progress during the second season registering about 18.2 %, an increase of 10 % with in a short period (Table 6).

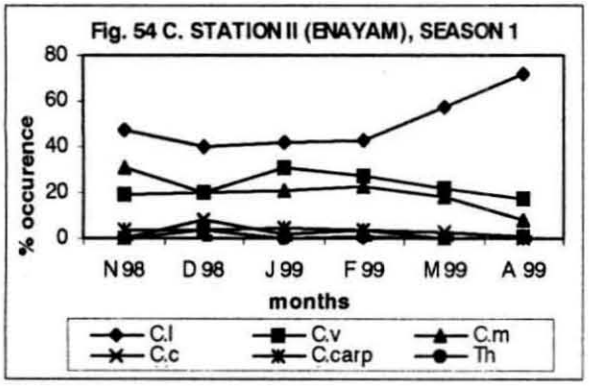
For a better understanding of the species competition between and within the species groups (ie. new invaders and conventional species) the data collected on monthly basis were analysed and the results are given in Fig. 54 E & F. During the first season the mussel fishing was rather erratic and samples were not available for certain months. *C. lobata* was totally absent in the former months and the competition noted

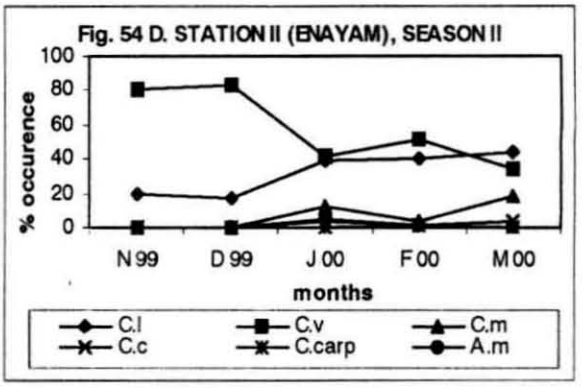
Table 6. Season- wise species composition at Colachel (Station III)

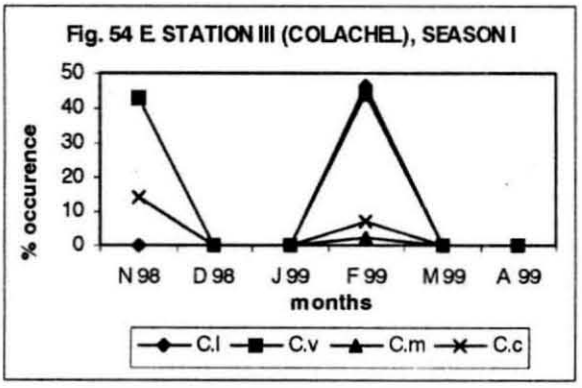
Sl. No.	Species	1 st season composition (%)	11 nd season composition (%)
1	*Cliona lobata*	40.0	31.8
2	*Cliona vastifica*	44.0	45.5
3	*Cliona margaritifera*	8.0	4.5
4	*Cliona celata*	8.0	18.2

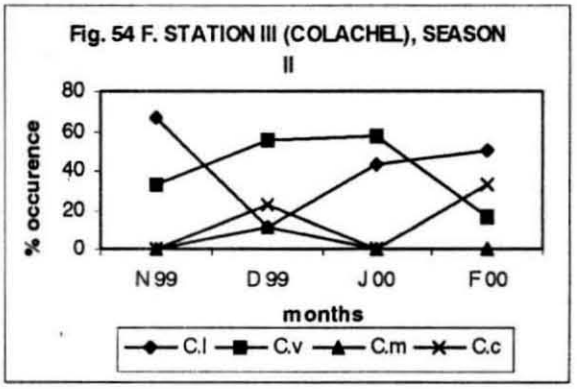












Figs. 54 A - F. Species -wise infestation pattern at Stations I-III
C. l: *Cliona lobata*; **C. v**; *Cliona vastifica*; **C. m**: *Cliona margaritifera*;
C. c: *Cliona celata*; **C. carp**: *Cliona carpenteri*;
Th: *Thoosa armata*; **Al**: *Alectona millari*; **A. m**: *Aka minuta*

was mainly between *C. vastifica* and *C. margaritifera*. But by February the condition changed and *C. lobata* took the lead with 46.5 % followed by *C. vastifica* with 44.2 % in species composition. *C. margaritifera* recorded only 2.3 % (species compositon). During the second season *C. lobata* could be seen dominating for two months (November and February) and in these months *C. vastifica* occupied only the second and third positions respectively in species composition. In the months of December (1999) and January (2000) *C. vastifica* dominated and *C. lobata* occupied only the third and second positions respectively. During November, 1999 and January, 2000 only two boring species (*C. lobata* and *C. vastifica*) could be seen in these beds. *C. margaritifera* was present only in December 1999 (11.1 %) and *C. celata* in December, 1999 and February, 2000 (22.2 and 33.3 % respectively) (Figs. 54 E & F).

Station IV, Kadiyapatnam (Map 1)

At Kadiyapatnam a total of 5 boring species could be collected as pests of brown mussel and they were present in both seasons alike (Map 3). Except two new invaders, (*C. margaritifera* and *C. lobata*) all the others (viz. *C. vastifica*, *C. celata* and *C. carpenteri*) were conventional species. *C. carpenteri*, though a conventional species in the Indian molluscan beds it is first reported from mussel at this centre and also from station II (Enayam) during the present study.

Here also, as in other stations, there is no information on boring sponge infestation on brown mussel in the past and as such it is difficult to make any comparative assessment of their incidence pattern or percentage species composition.

As seen from the collections, the incidence of boring sponges at this centre is quite negligible, 8 % during the first season (November, 1998 to February, 1999) and 13 % in the second season (November, 1999 to April, 2000) (Map 2). Here also, as in station III (Colachel), the conventional species *C. vastifica* dominated during both the seasons with a species compositon of 52.5 % and 47.4 % respectively (Table

7) indicating that *C. vastifica* is still dominating in these brown mussel beds though there is severe competition from the two new invaders, especially from *C. lobata*. *C. lobata* occupied the second position in species composition with 30 % and 37.2 % respectively in the first and second seasons. *C. celata* (conventional species) registered a species composition of 7.5 % in the first season, but the same registered a dip by the next season (2.6 %). *C. carpenteri* (conventional species) made its appearance in the first season with a species compositon of 2.5 %, but totally disappeared in the second season from these beds. *C. margaritifera* (new invader) recorded species composition of 7.5 % in the first season and the same increased to 12.8 % by the second season.

In order to collect a clearer picture of the competition among the two new invaders and the conventional species, the data collected on monthly basis were analysed separately and the same is given in Figs. 55 A & B. Here also the fishery was erratic due to inclement weather conditions

At this station *C. vastifica* dominated during the first season followed by *C. lobata*. *C. margaritifera*, *C. celata* and *C. carpenteri* together accounted for a total composition of 17.5 %. In the second season *C. lobata* dominated during January, 2000 and February, 2000 while in all the other months *C. vastifica* dominated. *C. margaritifera* was somewhat well represented in all the months and occupied the third rank in percentage composition. *C. celata* was present only during January, 2000 (Figs. 55 A & B). Competition between *C. lobata* and *C. vastifica* was well marked at this station also.

Station V, Cape Comorin (=Kanyakumari) (Map 1)

The monthly collections made from this centre (Station V) show that the boring sponge infestation was nil during the first season (October, 1998 to March, 1999) and hence the sampling for the next season was discontinued. The total length of mussel varied from 10 mm to 80 mm, and the various size groups collected are in full

Table 7. Season- wise species composition at Kadiyapatnam (Station IV)

Sl. No.	Species	1 st season composition (%)	11 nd season composition (%)
1	*Cliona lobata*	30.0	37.2
2	*Cliona vastifica*	52.5	47.4
3	*Cliona margaritifera*	7.5	12.8
4	*Cliona celata*	7.5	2.6
5	*Cliona carpenteri*	2.5	0

agreement with those collected from other centres during the present study.

Station VI, Mulloor (Map 1)

Mussel samples were collected from this centre (Station VI, Map 1) during October, 1999 to March, 2000. The incidence, at this centre was only 11.5 % (Map 2) while the same at Vizhinjam (Station I), a station quite close by, was 13.16 % for the corresponding period (second season).

Studies on species composition (from pooled data) indicated that *C. vastifica* (conventional species) accounted for the maximum infestation (49.3) followed by *C. lobata* (new invader) with a compositon of 43.5 %. (But at Vizhinjam, ie. Station I, the composition was just reverse during the same period, *C. lobata* ranked first and *C. vastifica* ranked second). The other two species *C. margaritifera* and *C. celata* registered poor composition at Mulloor; the former accounted for 4.3 % while the latter 2.9 % (Table 8). Earlier studies made by Thomas *et al.*, (1993) revealed that Mulloor mussel beds were infested with three species of sponges. *C. lobata*, *C. margaritifera* and *C. vastifica* during 1980 to 1982 period and *C. lobata* formed the dominant species ie., 60 %, followed by *C. vastifica* (20 %) and *C. margaritifera* (20 %).

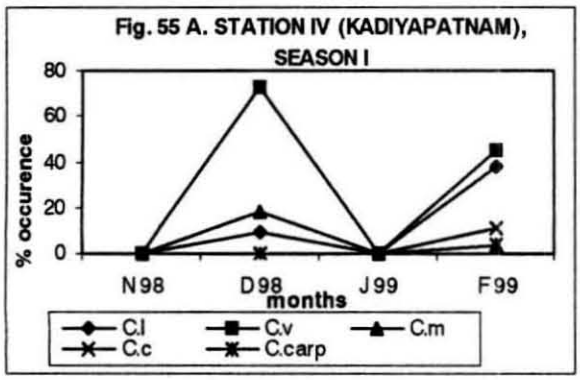
Out of six months for which investigations were made (October, 1999 to March, 2000), the dominance of *C. lobata* could be noted only during one month ie. November, (1999) while in all other months, *C. vastifica* dominated. *C. margaritifera* was present during three months; November, 1999, February, 2000 and March, 2000, while *C. celata* in two months (November, 1999 and February, 2000) (Fig. 55 C).

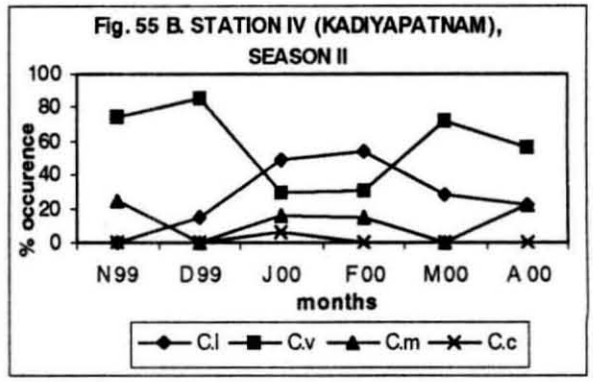
Statistical analyses

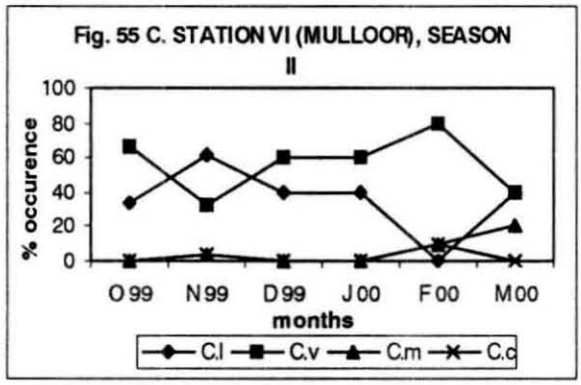
The various size-groups were classified into three class intervals; 0-45 mm, 45-90 mm and 90-135 mm for analysis. Two-way ANOVA was performed for total as well as sponge infested mussel population and species of boring sponges. A total of

Table 8. Distribution of various species at Mulloor (Station VI)

Sl. No.	Species	October, 1999 to March, 2000 (%)
1	*Cliona lobata*	43.5
2	*Cliona vastifica*	49.3
3	*Cliona margaritifera*	4.3
4	*Cliona celata*	2.9







Figs. 55 A - C. Species-wise infestation pattern at Stations IV & VI
C. l: *Cliona lobata*; **C. v**; *Cliona vastifica*; **C. m**: *Cliona margaritifera*; **C. c**: *Cliona celata*; **C. carp**: *Cliona carpenteri*

four stations, ten months, three size groups and eight species were compared by two-way analysis.

In the first case frequencies of the total brown mussel population were compared with factors viz. stations, months and size groups. Significant difference was observed between size groups (P<: 0.01). No significant difference was noted between stations and months (P > 0.05). Significant interaction effect was observed between stations and size groups (P<: 0.01) and between months and size groups (P <: 0.01). But between sampling stations and months (P> 0.05) no interaction effect was observed (Table 9 A-C).

Similarly frequencies of sponge infested brown mussel population were compared between stations, months and size groups. Significant variation was observed between stations and size groups at 1 % level where as no significant difference was noted between months (P > 0.05). Interaction effect between size groups and months and also between stations and months showed no significance. But between months and size groups, variation between seasons was highly significant (P <: 0.01) (Table 10 A-C).

Comparing stations, species of boring sponges and months, significant variation was observed between stations and species (P <: 0.01). Between months no significant difference was observed. Interaction effect between stations and species showed significance at 1 % level. But no interaction effect was observed between months and stations and between species and months (P > 0.05) (Table 11 A-C).

4. 3. BIODIVERSITY STUDIES

4.3.1. Of sponge infesting mussels

The Porifera is one among the rare groups of animals on which the

Table 9 A-C. Analysis of variance for frequencies of total brown mussel population

9. A

Source	SS	df	MS	F	P	Remarks
A	23.704	3	7.901	0. 005	0.999	NS
B	30.689	9	3.410	0. 002	1. 000	NS
AB	118.652	27	4.395	0. 003	1. 000	NS
E	116855.485	80	1460.694			

A- Station, B- Month, AB-Station and month, E- Error, NS - not significant

9. B

Source	SS	df	MS	F	P	Remarks
A	23.704	3	7.901	0. 042	0.989	NS
C	88571.328	2	44285.664	233.131	0.000	***
AC	7917.781	6	1319.630	6.947	0. 000	***
E	20515.717	108	189.960			

A- Station, C- size group, AC-Station and size group, E- Error, NS - not significant, *** - Highly significant

9. C

Source	SS	df	MS	F	P	Remarks
C	88571.328	2	44285.664	209.133	0.000	***
B	30.689	9	3.410	0. 016	1.000	NS
BC	9368.273	18	520.458	2.458	0.003	**
E	19058.273	90	211.759			

B- Month, C- size group, BC- Month and size group, E- Error, NS - not significant, *** - Highly significant, ** - Significant at 1 % level

Table 10. A-C Analysis of variance for frequency of brown mussel population infested by boring sponges

10. A

Source	SS	df	MS	F	P	Remarks
A	3348. 076	3	1116. 025	5.989	0.001	***
B	1274.709	9	141.634	0.760	0.653	NS
AB	2852.523	27	105.649	0.567	0.951	NS
E	14908. 032	80	186.350			

A - Station, B - Month, AB-Station and month, E- Error *** - Highly significant, NS - Not significant

10. B

Source	SS	df	MS	F	P	Remarks
A	3348. 076	3	1116. 025	11.870	0.000	***
C	6238.543	2	3119.272	33.178	0.000	***
AC	2642.822	6	440.470	4.685	0.000	***
E	10153.899	108	94. 018			

A - Station, C - Size group, AC- Station and size group, E- Error, NS - Not significant, *** - Highly significant

10. C

Source	SS	df	MS	F	P	Remarks
C	6238.543	2	3119.272	22.739	0.000	***
B	1274.709	9	141.634	1. 032	0.421	NS
BC	2524. 069	18	140.226	1. 022	0.444	NS
E	12346. 019	90	137.178			

C - Size group, B - Month, BC- Month and size group, *** - Highly significant, NS - Not significant

Table 11. A-C. Analysis of variance for species of boring sponges

11. A

Source	SS	df	MS	F	P	Remarks
A	2818.940	3	939.647	16. 034	0.000	***
B	819.634	9	91. 070	1.554	0.129	NS
AB	1700.666	27	62.988	1. 075	0.369	NS
E	16408.453	280	58.602			

A- Station, B- Month, AB-Station and month, E- Error, *** - Highly significant, NS - not significant

11. B

Source	SS	df	MS	F	P	Remarks
A	2818.940	3	939.647	34.662	0.000	***
C	8461. 086	7	1208.727	44.588	0.000	***
AC	2660. 347	21	126.683	4.673	0.000	***
E	7807.320	288	27.109			

A- Station, C- Species, AC-Station and species, E- Error, *** - Highly significant

11. C

Source	SS	df	MS	F	P	Remarks
B	819.634	9	91.7	1.946	0. 046	NS
C	8461. 086	7	1208.727	25.823	0. 000	***
BC	1232.906	63	19.570	0.418	0. 000	***
E	11234. 067	240	46.809			

B- Month, C- Species, BC-Month and species, E- Error, NS - not significant, *** - Highly significant

biodiversity data have hardly been documented. In the case of this group, species Information and related data are scanty. Therefore universal documentation tools and active communication among taxonomists working in different areas and related fields are highly imperative. Taxonomic and distributional data are essential for the study of sponge biodiversity. Modern multimedia techniques can be applied for biodiversity studies. Computer- based Biodiversity information system has been developed as a universal tool for biodiversity documentation. The Linneaus II software developed by ETI (Expert Centre for Taxonomic Identification) provides a helping hand for the documentation of biodiversity data on sponges (Rob, *et al.*, 1996).

As per the reports of the Zoological survey of India the Indian invertebrate fauna comprises 89,451 species. Some groups of invertebrates have been well documented in the past while others are not. Only very few studies have been made on the taxonomy and biodiversity of rare groups especially the marine sponges. As India is a signatory to the GATT and Biodiversity Convention (BDC), it is mandatory on our part to study, document and utilize our biodiversity wealth for the betterment of humanity.

The following statistical methods were used in the present study:

1. **Species richness index** (Margalef, 1957)
2. **Simpson's Index or species concentration factor** (Simpson, 1949)
3. **Shannon Weaver diversity index** (Shannon and Weaver, 1963)
4. **Heips evenness index** (Heip, 1974)
5. **Species dominance index** (Pielou, 1971)

Values of five diversity indices calculated for each sample are given in Table 12 A & B. Number of species (*s*) is defined as the total number of species encountered at the station. The number of species at the stations covered ranges from 4 (Station I) to 8 (Station II) (Map 3).

Table 12. A. Biodiversity indices

Station	Margalef Index (*d*)	Simpson's index (*Sp*)	Shannon Index *H (S)*	Pielou's index (*D*)	Heip's Index (*E*)	No. of Species (s)
1	0.6346	0.9990	1.6191	1.1680	1.349	4
2	1.1238	0.9997	1.8537	0.8914	0.769	8
3	0.9353	0.9960	1.6280	1.0115	1.023	5
4	0.8384	0.9985	1.8282	1.1359	1.305	5

Table 12. B. Mean (x), standard deviation (σ) and coefficient of variation (CV) of biodiversity indices

	Margalef's index (*d*)	Simpson's index (*Sp*)	Shannon diversity index *H (S)*	Pielou's index (*D*)	Heip's evenness index (*E*)
X	0.883	0.998	1.732	1.051	1.111
SD	0.176	0.001	0.109	0.109	0.233
CV	19.972	0.139	6.299	10.400	21.050

Pielou's (Pielou, 1971) index (*D*) is concerned with the dominance of species. At the stations covered, it is the highest at Station I (1.17) and the lowest at station 2 (0.89). Species dominance is inversely related to the number of species. The mean (x), standard deviation (σ) and the coefficient of variation (%) of this index in this study being 1.05, 0.11 and 10.4 correspondingly.

Simpson's (Simpson, 1949) index (*Sp*) of the degree of concentration or diversity is almost uniform for the Stations I, II and IV (0.999) covered, but it is relatively low at Station III (*Sp*= 0.996). Mean of this index is 0.998, standard deviation and coefficient of variation being 0.001 and 0.14 respectively.

Margalef's (Margalef, 1957) index (*d*) varies from 0.63 (Station 1) to 1.12 (Station 2), mean, standard deviation and coefficient of variation being 0.88, 0.18 and 19.97 respectively.

Shannon Weaver function (Shannon and Weaver, 1963), *H* (*S*), is the highest at Station II (1.85), showing maximum number of species (s= 8), while it is lowest at Station 1 (1.62) with the least number of species (s= 4). Mean (x), standard deviation and coefficient of variation for this index are 1.73, 0.11 and 6.30 respectively.

Heip's (Heip, 1974) index measures evenness of occurrence of species in a population. In the present study, it varies from 0.77 to 1.35. Low equitability indicates dominance of a few species, while high equitability reflects uniform distribution of species. Mean of this is 1.11, standard deviation and coefficient of variation being 0.23 and 21.05 respectively.

In conclusion, this study suggests that species number (*s*), Shannon-Weaver function, *H (S)* and Margalef's index (*d*) are the lowest at Station 1 (*s*= 4, *H* (*S*)

= 1.62 and *d*= 0.63) and the highest number of species and the greatest diversity at Station II (*s* = 8, *H* (*S*)= 1.85 and *d*= 1.12).

4. DISCUSSION

Soon after the appearance of the two invaders (*C. margaritifera* and *C. lobata*) on the pearl culture rafts at Vizhinjam in 1980, there was an effective spreading of the above two species to the natural beds of mussels, rock oysters, pearl oysters, *Thais rudolphi*, sacred chank etc. in and around Vizhinjam. The migration of the above species of boring sponges to the nearby molluscan beds was traced out for a period upto 1986 (Thomas *et al.*, 1983). The following observations were made above survey.

1. There was a sudden spurt in the general incidence and infestation pattern in every bed either through the activity of the two new invaders or through triggering the activities of the other conventional species in the beds.

2. The initial spurt in their incidence generally subsided in natural beds and reached a level very close to that seen prior to the invasion (of *C. margaritifera* and *C. lobata*). But this was not seen in the culture systems maintained at Vizhinjam.

3. There has been severe competition between the two new invaders and in this struggle, *C. margaritifera* proved to be more adaptable and hence more successful in the molluscan culture systems.

4. In the competition between conventional species, *C. celata* took the lead in every bed.

The above situation changed drastically during the present study made in 1999 to 2001, after a long spell of about 21 years, utilizing specimens from different natural mussel beds and also from culture systems at Vizhinjam (Arabian Sea), Ashtamudi Lake (west coast) and Tuticorin (Gulf of Mannar). Salient features emerged during the present study may be summarised as follows:

1. Both *C. lobata* and *C. margaritifera* are still widely distributed all along the coast but the former is dominating in different beds also in many centres. *C. margaritifera* infestation at present is found to be quite negligible.

2. The conventional species, *C. vastifica*, compete with *C. lobata* (new invader) in all beds and occupy the second position.

3. *C. celata* infestation is found negligible in the various mussel beds.

4. The infestation or incidence at present is not quite severe except at Enayam. The lowest infestation was found at Colachel during the second season (5.5 %), at Kadiyapatnam during the first season (8%), and at Vizhinjam during the first season (9.28 %). This shows that the infestation is slowly getting decreased in the various beds and ultimately it may reach a level of 5 to 8 % as seen prior to the invasion of the two new invaders (*C. margaritifera* and *C. lobata*) in 1980.

5. On mussel culture rafts at Vizhinjam, *C. lobata* formed the dominant species (upto 61 %) followed by *C. vastifica* (26 %). *C. margaritifera* and *C. celata* together account for 13 % only. The incidence of boring sponges noted on these rafts is 23 %. This, when compared with the same in natural mussel beds off Vizhinjam, is quite high (the present study revealed that the incidence is 9.8 % and 13.16 % respectively in the first and second seasons). On mussel culture systems at Vizhinjam

an incidence as high as 47 %, 60 % and 80 % could be noticed during 1980, 1981 and 1982 respectively (Thomas *et al.*, 1993).

6. Out of the six stations studied Enayam (Station II) is the only centre where a high incidence could be noticed (63.16 % and 42.6 % respectively during first and second seasons). The number of boring species recorded from this centre also totalled to 8. During the first season, *C. lobata* ranked first in species composition (52 %), while *C. vastifica* (44.6 %) ranked first during the second season (pooled data) (Table 5).

7. Taking the species composition into consideration it may be stated that *C. vastifica* is more adaptable than any other conventional species and *C. lobata* (the new invader) is in severe competition with *C. vastifica* in gaining dominance. *C. margaritifera* became less vigorous in its activity when compared with *C. lobata* during the last twenty-year period (from 1980 to 2000).

8. In the estuarine systems (Ashtamudi Lake) *C. vastifica* is the only species infesting the molluscan shells.

To assess the performance of the two new invaders in relation to conventional species present in the various mussel beds the data collected from all the centres for two seasons were pooled and the species composition and incidence were calculated. Out of 5, 600 shells examined 995 were infested by boring species and this works out to a incidence of 17.76 %. In various natural beds off Mulloor and Kovalam there was a sudden hike in the species composition of boring species soon after the appearance of the two new invaders in 1980 when it went upto 54 % and 48 % respectively at Mulloor and Kovalam. But this sudden spurt came down gradually in these beds. The present incidence of 17.76 % noted from various beds investigated under the present study is somewhat high. The activities of *C. margaritifera*, one of

the two new invaders, though subsided considerably, the activities of a conventional species, *C. vastifica*, showed considerable increase in the southwest coast in general, and this hike in infestation may be attributed to the hike in the activity of *C. vastifica* in the various beds.

Pooling the data from all the six centres for both seasons, the species composition of various species was calculated and it could be seen that *C. lobata* (new invader) ranked first with a composition of 44.6 % followed by the conventional species, C. *vastifica*, with 36.7 %. *C. margaritifera*, the second among new invaders, occupied only the third position with 12.5 % (Table 13). All the other species, put together accounted for a species composition of 6.2 % only.

For getting a clearer picture on the performance of the above species, the data collected from different stations for each season were pooled. It could be seen that *C. lobata* dominated in the first season with a species composition of 47.4 % while *C. vastifica* dominated during the second season with a species composition of 44 %. *C. margaritifera* occupied the third position with a species composition of 15.6 % and 9.0 % respectively during the first and second seasons. All the other species had only very low representation during the period of the present study (Table 14).

Month-wise trend of dominance of species showed that out of all samples collected over a period of 40 months from different centres, 23 samples showed the dominance of *C. vastifica*. This clearly shows that *C. vastifica* is more active at present and *C. lobata* may be considered the next in dominance in the various molluscan beds studied at present.

Table 13. Species composition for all stations for both seasons (pooled data)

Sl. No.	Species	Species composition (%)
1	*Cliona lobata*	44.6
2	*Cliona vastifica*	36.7
3	*Cliona margaritifera*	12.5
4	*Cliona celata*	4.1
5	*Cliona carpenteri*	1.0
6	*Thoosa spp.*	0.5
7	*Alectona millari*	0.4
8	*Aka minuta*	0.2

Table 14. Species composition based on season-wise pooled data from different centres

Sl. No.	Species	I st season (%)	II nd season (%)
1	*Cliona lobata*	47.4	41.4
2	*Cliona vastifica*	30.3	44.0
3	*Cliona margaritifera*	15.6	9.0
4	*Cliona celata*	3.9	4.3
5	*Cliona carpenteri*	1.7	0.2
6	*Thoosa spp.*	0.7	0
7	*Alectona millari*	0.4	0
8	*Aka minuta*	0	1.1

5. STUDIES ON THE MIGRATION OF BORING SPONGES TO MOLLUSCAN CULTURE SYSTEMS

INTRODUCTION

The availability of suitable substratum for attachment is one of the factors which govern the distribution and abundance of sedentary organisms. In tropical waters, where most invertebrates have prolonged breeding cycle, the problem of availability of suitable space is often very acute, resulting in the overcrowding of several generations in a limited space. While many attach themselves to the substratum superficially, some bore into hard calcareous objects. Sponges of the family Clionidae are known to inflict adverse effects on molluscs as their calcareous shells are plenty, available and easily accessible for clionid larvae to settle and grow.

The total number of boring sponge species recorded from the Indian Seas till 1979 was 32 (Thomas, 1979 B) and this number, as compared to that in any oceans/ seas of the world is far high. This shows that the calcium carbonate-secreting animals in the Indian seas are under constant threat of sponge attack. The rate of infestation (ie. Infestation/ 100 shells) has always found to be less and well within the predictable limits (3-10 %) in the natural beds for conventional species of boring sponges year after year along the southeast and southwest coast of India. But this situation drastically changed by 1980 when two new invaders (*Cliona margaritifera* and *Cliona lobata*) made their appearance on the pearl oyster culture rafts moored at Vizhinjam Bay. It may be mentioned in this context that the incidence of boring sponges recorded from these rafts, just prior to the above invasion, was only 3-8 % (Appukuttan, 1987). But the incidence was found high during the subsequent season (47 % in 1980 and 60 % in 1981,Thomas *et al.*, 1993) among raft-cultured pearl oysters. Similarly the brown mussel collected from culture rafts moored at Vizhinjam indicated 8.2 % incidence in 1982, 10.6 % in 1983 and 20 % in 1984. These indicate a sudden spurt in the incidence pattern among tended stock of molluscs mainly due to the activity of the above mentioned two new invaders.

From the culture rafts, these two new invaders started spreading to the commercially important molluscan beds in and around Vizhinjam. During a subsequent

survey which ended in 1986, it could be seen that *C. margaritifera* migrated to the raft cultured pearl oysters at Tuticorin and *C. lobata* to the chank beds off Tiruchendur (both places on the southeast coast) within a period of two years (Thomas *et al.*, 1993).

It could also be noticed during the above survey that there was severe competition for space (here shell) between the conventional species (mainly, *C. celata*, *C. vastifica*, *C. carpenteri*), which were already present in these beds and the new invaders (*C. margaritifera* and *C. lobata*). Apart from such competition between these two groups there was severe competition among the various species within each group. In the between-group competition it was the new invaders (*C. margaritifera* and *C. lobata*), which won initially, but in the competition within the invader group it was *C. margaritifera*, which succeeded finally. In the competition among conventional species the one which has traditional dominance in that particular bed generally wins. When one species is directly inhibited by another, it might activate a third species to multiply disproportionately causing epidemics at times (Thomas, 1990).

It could be found during the earlier studies (Thomas, *et al.*, 1983) that the boring sponge *C. vastifica* had succeeded in colonizing the estuarine realms in Ashtamudi Lake near Kollam (= Quilon), Zuari and Mondovi estuaries of Goa due to its euryhaline nature. Hence this species (*C. vastifica*) poses a serious threat to any future molluscan farm in our estuaries.

In order to collect the information on incidence pattern, migration of boring sponges from the wild to the tended stocks etc., samples were collected from pearl culture rafts at Tuticorin (Gulf of Mannar), and brown mussel culture rafts at Vizhinjam (Arabian Sea). Since culture activities for brown mussel were not in vogue in any of the estuaries, some attempts were made to cultivate them in Ashtamudi Lake (Dalavapuram) in Kollam district (Map 1) for collecting details on boring sponges and their infestation pattern in an estuarine system.

5. 1. Raft cultured mussel at Vizhinjam

1. a. MATERIAL AND METHODS

In order to compare and contrast the rate of sponge infestation (incidence %) in natural beds with that in culture systems, a study was taken up at Vizhinjam utilizing the brown mussels reared through rope-culture methods. One hundred specimens taken form the raft at random were examined for sponge infestation, and the salient findings emerged are summarized below:

1. b. Hydrographic conditions of the culture site

Water samples were collected from the culture site at monthly intervals for a period of six months (October, 1998 to March, 1999). Dissolved oxygen was determined by standard Winkler method. For the estimation of salinity, samples were collected in polythene bottles and estimated titrimetrically by the Mohr Knudsen method. Inorganic phosphate, nitrate and silicate were estimated following the method given by Strickland and Parsons (1960).

1. c. RESULTS

The surface temperature varied from a minimum of 31 º C to a maximum of 33 º C. The salinity ranged from 30.38 ppt to 35.4 ppt. Dissolved oxygen values varied from a minimum of 3.26 ml/l in March to a maximum of 5.11 ml/l in January. Maximum nitrate content was recorded in March (1.58 µg at/l) and the lowest in February (0.77 µg at/l). Phosphate content fluctuated between 0.509 and 2.916 µg at/l during the study period whereas silicate content varied from 0.687 to 1.39 µg at/l (Fig. 56).

During the present study the incidence of boring sponges noted was 23 %, which was quite high as against the same in natural beds (9.28 %). The infestation usually starts when the mussel is 35 mm length and may be seen in different size-

groups upto 70 mm (Fig. 59). Maximum infestation could be seen in 50-55 mm size-group with the right valve getting infested mostly.

Four species of boring sponges were found to occur on the cultured brown mussels. *C. lobata* formed the dominant species (61 %), followed by *C. vastifica* (26 %), *C. margaritifera* (9 %) and *C. celata* (4 %) (Fig. 58). As mentioned earlier, in the competition between the two new invaders (*C. margaritifera* and *C. lobata*) it is *C. margaritifera* which wins finally but in the present observation both were found suppressed by *C. vastifica* which is a widely distributed conventional species in the Indian molluscan beds (Thomas, 1979 B).

5. 2. Rope- cultured brown mussel at Dalavapuram

2. a. MATERIAL AND METHODS

In order to study the boring sponge infestation in an estuary a site (Dalavapuram) in the Ashtamudi Lake (Map I) was selected. Seeds of brown mussel were collected from the intertidal areas of Neendakara harbour (Kollam, Arabian sea) then cleared off foulers, silt etc. and seeded on to nylon ropes (1 m length and 10 mm diameter) and wrapped with cotton mosquito netting. Seeds of 35 mm average size were used in the experiment. Seeded ropes were suspended from fixed cassuarina poles (Plate 6). The mussels got attached firmly to the nylon rope by means of freshly secreted byssus threads and the mosquito netting disintegrated fully within a period of 10 days. The cultured mussels were examined regularly for a period of 150 days (from December, 1998 to May, 1999).

2. b. Hydrographic conditions

Temperature, salinity, dissolved oxygen, nitrate, phosphate and silicate were monitored throughout the culture period.

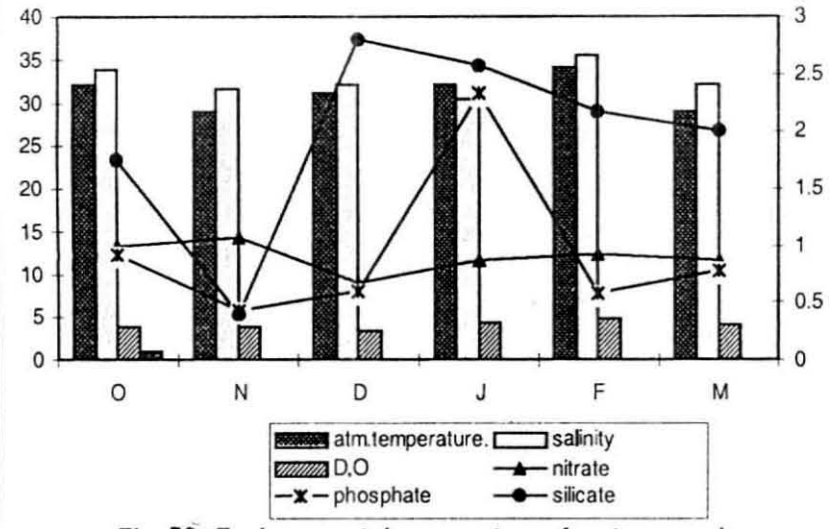


Fig. 56 Environmental parameters of water samples collected from mussel culture site at Vizhinjam

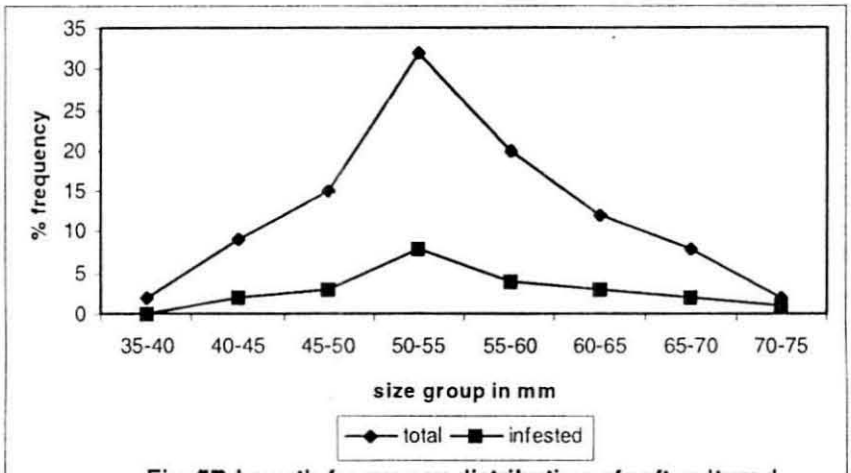


Fig. 57 Length frequency distribution of raft cultured brown mussel at Vizhinjam

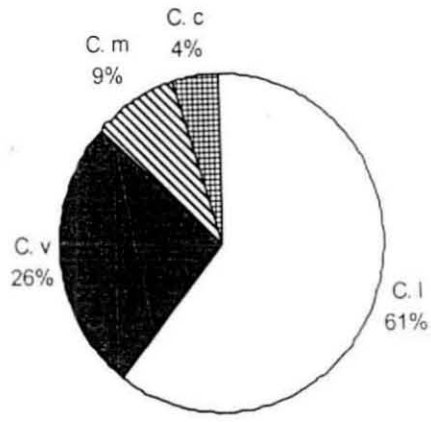


Fig.**58** species composition of boring sponges at Vizhinjam culture raft

C. l: *Cliona lobata*; **C. v**; *Cliona vastifica*; **C. m**: *Cliona margaritifera*; **C. c**: *Cliona celata*

2. c. RESULTS

Dissolved oxygen values varied from 3.26 ml/l to 5.51 ml/l and temperature values were within the normal range, ie. 30° C to 33° C. The salinity ranged from 26.49 ppt to 31.8 ppt. Nitrate concentration varied from 0.77 μg at/l (February) to 1.58 μg at/l (March), phosphate from 0.50 μg at/l (December) to 2.91 μg at/l (March) and silicate from 0.68 μg at/l (December) to 1.39 μg at/l (February) (Fig. 59).

Monitoring of the cultured mussels was done regularly for the growth and survival. Mussel seed with an average size of 31.5 mm (length) in December reached to an average size of 56.52 mm (length) within a period of 150 days or by May, 1999.

Out of 100 specimens of mussels examined, 18 % were infested with boring sponge *C. vastifica* and the size group of 45-50 mm registered the maximum infestation (Fig. 60). The openings made by the sponge were confined to the umbo region and the right valve was found infested mostly. No other species of boring sponge was found at this station.

DISCUSSION

Hartman (1958) opined that *C. vastifica* is a more adaptive and plastic species being capable of inhabiting a wide variety of habitiats in regard to depth, reduced salinity and exposure to air than is true of *C. celata*, and in Indian waters *C. vastifica* is more dominant than *C. celata* on oyster beds. "Populations of *C. vastifica* have invaded brackish water in a number of regions of the world and have undergone parallel morphological changes in each area. Three populations along the American Atlantic coast apparently merit specific rank " (Hartman, 1958).

In India, *C. vastifica* is commonly found in lakes and brackishwater areas and is reported from Chilka Lake, Adayar River and Ennore backwaters (Annandale,

Plate 4. View of mussel culture on ropes suspended from cassuarina poles in Ashtamudi Lake

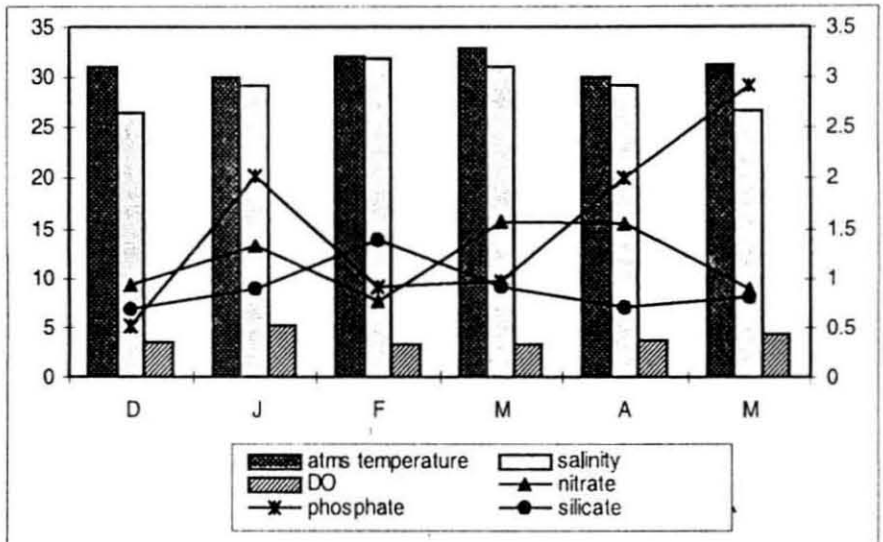


Fig. 59 Environmental parameters of water samples collected from mussel culture site in Ashtamudi Lake

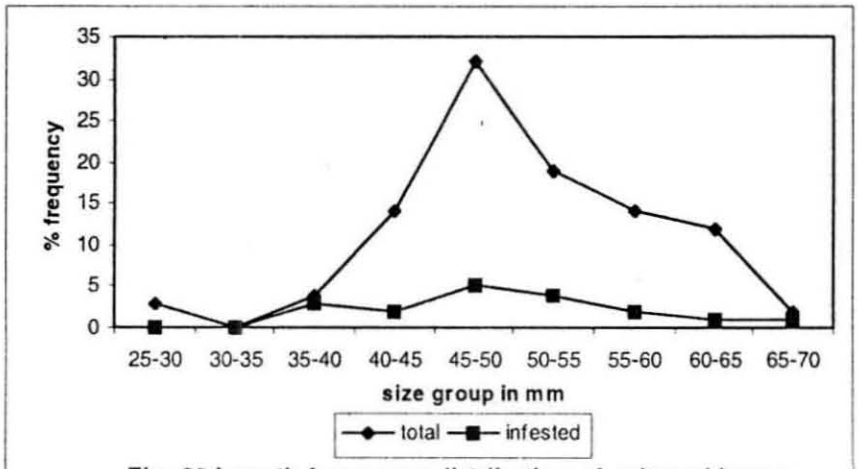


Fig. 60 Length frequency distribution of cultured brown mussel at Dalavapuram

1915),Zuari and Mandovi estuaries, Goa (Thomas, 1975) and Ashtamudi Lake (Thomas *et al.*, 1983). The extreme salinity tolerance, capacity to form gemmules and faster growth are the three factors which made *C. vastifica* a successful species in the estuarine systems (Thomas, 1975).

6. PATHOLOGICAL STUDIES

6 . 1 Pathological aspects of sponge boring

1. INTRODUCTION

When a boring sponge attacks a live mollusc, the latter may react to the intruder in different ways. This may produce considerable physical as well as physiological strain on the host. When the inroads of boring sponge fill the interior of shell, some of the chambers found close to the nacreous layer may put forth papillae piercing the nacreous layer (Fig. 2). When diverging spicules of those papillae touch the mantle epithelium it may produce much irritation or damage to the soft mantle epithelium.

The boring sponge etches out minute particles of calcium carbonate from the interior of the shell and thus forms chambers and canals inside (Fig. 25). In advanced stages the shell becomes brittle due to the dense proliferation of chambers and canals inside (Fig. 32). In the case of thick shells, (especially of the rock oyster or of the sacred chank, *Xancus pyrum*), the chambers formed by the sponge usually unite together forming a continuous cavity inside the shell with only the outer and inner layers (periostracum and nacreous layer respectively) intact (Fig. 9 A, B). Such shells with continuous cavities inside may crumble at the slightest pressure. This type of shells are often described as "fragile shells" or "spongy shells".

As the inroads of sponge becomes extensive inside the shell, more and more calcareous chips are expelled from the interior of the shell, and this may result in a gradual loss of weight on the part of the host.

In the initial stage of infestation small pores are seen at the outer part of the umbo region of the shell. But, as the sponge spreads through the interior of the shell, such pores may be seen scattered all over the surface of shell. The excurrent and incurrent papillae of the sponge are protruded through these pores found on the surface. In more advanced stages of infestation when the "sponge mass" increase considerably inside the shell, more water is needed and this is made possible by

piercing the inner nacreous layer of the shell and drawing water from the space in between the shell and mantle of the mollusc. These papillae made by the sponge, when expanded, may touch the mantle epithelium of the mollusc giving perpetual irritation to the host. The presence of minute pores at the surface of the molluscan shell is so characteristic of any boring sponge that this manifestation is often termed "porosis". When one side (usually the upper) is beset with pores, it may be called monofacial porosis, and when while both surfaces (inner and outer) are beset with pores it may be termed bifacial porosis (Thomas, 1983).

When openings made by the sponge inside the shell (nacreous layer) are repaired by nacreous material of the live mollusc, a black patch is often formed at the site of the original pore. By such constant repairs a blister is formed at this spot and such blisters often contain a pigmented summit (Figs. 11, 19, 22 B, C, D, 26 C, 33 B, C, D). How larger areas with pigment (or plate-like pigment) are formed is shown in Fig. 33 D and explained under *C. margaritifera*. Such pigment formation inside the shell is often denoted by the term melanosis (Thomas, 1983).

When the openings made by the boring sponge are concentrated in the nacreous layer it is often noted that the nacreous material becomes less lustrous and that in advanced stage this area may become very rough due to the poor coating of nacreous material. The total damage of the nacreous secreting cells on the mantle surface is the main reason for this disease (nacreerosis).

The above-mentioned are some of the physical effects of sponge infestation. A detailed account of the various physical stress situations noticed in other shells is given in Thomas, (1983) and about 12 different diseases have already been documented by him.

2. MATERIAL AND METHODS

During the present study, bored shells of *Perna indica* collected off the southwest coast were examined and the various pathological manifestations were recorded in detail.

3. RESULTS

Out of a total of 5, 600 shells examined during the present study 995 (or 17.8 %) were found infested with boring sponges. The various pathological manifestations (physical) were tabulated and the results are given below in a tabular from (Table 15).

All specimens infested with sponges exhibited monofacial porosis. The concentration of surface pores decrease from umbo region to the marginal zones of the shell. Bifacial porosis could be noted only in 19.1 % of infested shells. All severely infested shells (8.2 %) were fragile. Blisters were noted in 46.3 % of the infested shells. Discoloration, nacreous erosion and melanosis could be noticed only in the advanced stages of infestation (Plate 5). These results indicate that in brown mussel only six pathological manifestations are generally seen in hard parts.

4. DISCUSSION

The extent of damage done to the live molluscan shell by boring sponge cannot be assessed by superficial examination of the shell alone, as the surface of the shell may have only minute openings which may be distributed sparingly. Hence it is necessary to slice the shell vertically and horizontally and examine it carefully for assessing the magnitude of damage caused to the shell. Cavities made by sponge inside the shell would make the shell fragile and weak and such shells become susceptible to further damage by secondary invaders and borers like polychaetes, bacteria and fungi (Alagarswami & Chellam, 1976). It is often noticed that the mantle of such shells would become flabby followed by the formation of dark pigmented areas (pustulosis) exactly opposite to the holes made by sponge at the inner aspects of the

Table 15. Pathological manifestations of sponge infested mussel shells

Sl. No.	Features	No. of shells	%
1	Blister formation	466	46.3
2	Discolouration	197	19.8
3	Melanosis	130	13.1
4	Bifacial porosis	190	19.1
5	Fragility	82	8.2
6	Nacreerosis	76	7.6

Plate 5. A - E

A. Blisters with pores at the summit

B. Blister and discoloration

C. Pores, blisters and pigments distributed inside the umbo region of the shell

D. Bifacial porosis

E. Inner view of shell showing various types of blisters and openings formed inside (highly magnified view)

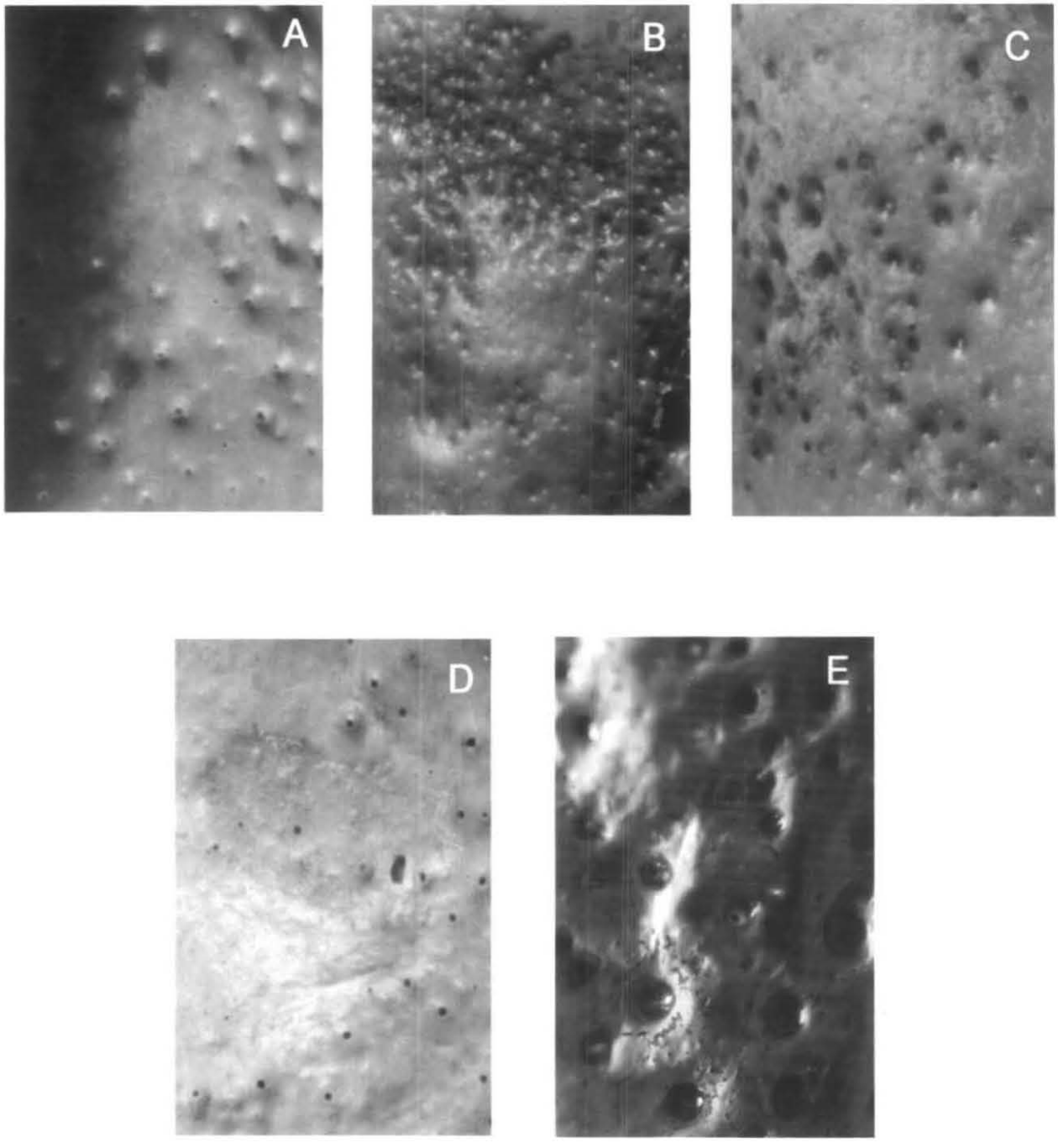


Plate 5. A-E. Pathological manifestations of sponge infested *Perna indica* shells

shell. This tissue often gets detached from the shell and presents a diseased look (Hornell, 1904). Inorder to study whether any biochemical change is effected in the soft tissue of the mollusc due to sponge infestation, some studies were taken up and the details, both cytological and histopathological, are furnished in the next section.

6. 2. Histological and transmission electron microscopic studies

1. INTRODUCTION

In view of the frequent occurrence of boring sponges in bivalves, histological studies were undertaken in *Perna indica* infested by various boring sponge species. The mantle epithelium plays a defensive role against the borer. It secretes excess of nacreous material to prevent the contact of sponge with the soft parts of the mollusc. The mantle epithelium just opposite to the pores through which the sponge papillae project out was separated carefully and this tissue was used in taking histological sections. Inorder to illustrate tissue level changes in mantle and adductor muscle sections of normal tissue were taken and compared with those of infested ones.

The ultrastructure details of the changes that occur in the outer surface of the mantle tissue during shell damage were also investigated. Sponge infested and uninfested mussels, obtained from the collection centres along the southwest coast, were examined and compared at ultrastructure level in regard to the condition of the mantle epithelium.

2. REVIEW OF LITERATURE

Gulka and Chang (1983) investigated on the prokaryotic infestation associated with the mass mortality of *Plactopecten magellanicus*. Dix (1973) highlighted the role of mantle and pearl sac in pearl formation of the pearl oyster

Pinctada maxima. The occurrence, prevalence, seasonality and histopathological progression of a cellular disorder in the mussel *Mytilus edulis* from Yaqiuna Bay were studied by Mix (1983). Balouet *et al.*, (1983) discussed the possible relationship between the outbreak of haemocytic parasitosis and the possible relationship with other parasites. Microscopic anatomy of the mantle of the pearl oyster *Pinctada mazatlantica* was described by Garcia-Gasca *et al.*, (1994).

Kagoo and Ayyakkannu (1994) investigated on the pathological changes caused by parasites in the gastropod *Chicoreus ramosus*. Morrison (1993) described the structure of mantle and mantle lobes of the oyster *Crassostrea virginica* using both light and scanning electron microscopy. The edge of the mantle in earl oyster *Pinctada margaritifera* was studied from a morphological and histological point of view by Zahab *et al.*, (1992).

The organisation of the mantle edge and the fine structure of the mantle epithelia (dorsal mantle, mantle edge, mantle cavity) of the neopilinid limpet *Laevipilina antarctica*, was described by means of transmission electron microscopy (Schaffer and Haszprunar, 1997). Bubel (1973, 1973 A) described the fine structure of basal cells at the base of the periostracal groove of some marine bivalves. He also examined the cells lining the periostracum groove in some marine bivalves by means of electron microscopic methods. A further study of the cells of the inner face of the outer fold of the mussel *Mytilus edulis* was also undertaken during periostracum repair. According to Lee *et al.*, (1997) blister-like lesions on the foot of cultured abalone caused mortalities as high as 50 % to 60 % in at least three abalone culture facilities at the vicinity of Dalian, China. Zahab *et al.* (1992) studied the ultrastructure of the pearl oyster *Pinctada margaritifera*.

3. MATERIAL AND METHODS

Marginal zone of the mantle tissue and adductor muscle of the brown mussel were processed to study the histological manifestations of boring sponge

infestation. Tissues taken from uninfested mussel were used as control. The specimens (70-75 mm) were depurated for a period of 48 hours in 20-litre plastic troughs. They were then dissected out and the relevant tissues were fixed immediately in Bouin's fixative (ABF) for 24 hours. The tissues were then dehydrated in a graded alcohol series (50 %, 70 %, 80 %, 90 %, 95 %, and 100 %). Following dehydration, they were kept in xylene and left in a mixture of paraffin wax with a solidification point at 56-60 ° C. Blocks prepared were sectioned using a rotary hand-microtome. Sections of 7 µm thickness were taken at room temperature (25-28 °C). The sections were deparaffinised, dehydrated, and stained using haematoxylene and eosin (Preece, 1972). Photographs of areas showing gross pathological changes were taken using Nikon AFX-DX II camera and the results interpreted.

The processing of tissues for TEM study was done as per the method recommended by Robinson *et al.*, (1985). The tissues from the outer fold of the marginal zone of mantle were used for ultrastructure studies. Tissues from uninfested mussels were also processed in the same pattern for comparison.

The specimens used for the study were depurated for 24 hours in filtered seawater, tissues dissected out, trimmed into small bits and fixed in 3 % ice-cold glutaraldehyde for three hours. Three washes were given in phosphate buffer (pH 7.2) and post fixed in 1 % osmium tetroxide for two hours followed by three washes each of 15 minutes duration in fresh buffer to remove excess fixative. The tissues were washed in double distilled water three times, dehydrated in graded alcohol series of 30 % acetone at 4 ° C (50 %, 70 %, 80 %, 90%, 95% and 100%) for fifteen minutes. The samples were cleared in propylene oxide. Two changes of fifteen minutes duration each were given. Spurr-embedding media was used for infiltration. The embedded tissues were kept in an incubator at 70° C for 24 hours till polymerization completed. Ultra thin sections were taken using glass microtome. The sections were stained with lead citrate, dried, and observed under Hitachi 600 Philips CM 10 electron microscope. Desired areas were photographed and the results interpreted.

4. RESULTS

In order to ascertain the pathological changes in sponge-infested mussel tissues, the gross pathological changes were noted in mantle tissue and adductor muscle. Histopathological examinations of the infested tissues revealed the following changes in tissue morphology:

1. Mantle

The histological details of mantle revealed the presence of three zones, an outer marginal zone with three folds, a pallial zone and a central zone. A single layer of stratified columnar epithelial cells of 40-50 μm thick was present in the outer marginal zone. The inner and outer surfaces were characterised by numerous mucus cells. The inner epithelium possessed dense cilia, large secretory cells, melanin pigment and deeply stained basal ovoid nuclei.

In normal tissue the periostracum was visible as a thin yellowish fold. The periostracal shell material was secreted from the periostracal groove located in this area (Plate 8 A). The secretion of periostracal material was affected in sponge-infested tissues (Plate 8 B). In normal tissue only a few haemocytes were present in the marginal zone of mantle (Plate 8 C). Haemocytosis, ie. increase in the number of haemocytes was observed in infested tissues (Plate 8 D).

Other tissue anomalies included sloughing of the outer epithelial layer and increased secretion of wandering secretory cells (Plates 8 E, F). Another notable feature was vacuolisation in various degrees in the mantle epithelium.

2. Adductor muscle

The adductor muscle was characterised by two histologically distinct portions, an opaque portion consisting of smooth muscle cells, and a translucent portion of cross striated cells. The muscle fibres were enclosed by several facial sheaths or

Plate 6. A - F

A. Marginal zone of uninfested mantle (C.S, H & E, × 200) PE- pigmented epithelium, WS- wandering secretory cells, OF- outer fold, MF- middle fold, P- periostracum

B. Marginal zone of sponge infested mantle (C.S, H & E, ×200) OF- outer fold, MF- middle fold, PG- periostracal groove

C. Outer fold of uninfested mantle (OF) (C.S, H & E, ×200) H- haemocyte

D. Outer fold of sponge infested mantle (OF) (C.S, H & E, ×200) Note the Infiltration of haemocytes (H) (arrow)

E. Middle fold of uninfested mantle (MF) WS- wandering secretory cells (C.S, H & E, ×200)

F. Middle fold of sponge infested mantle (MF) WS- wandering secretory cells (C.S, H & E, ×200)

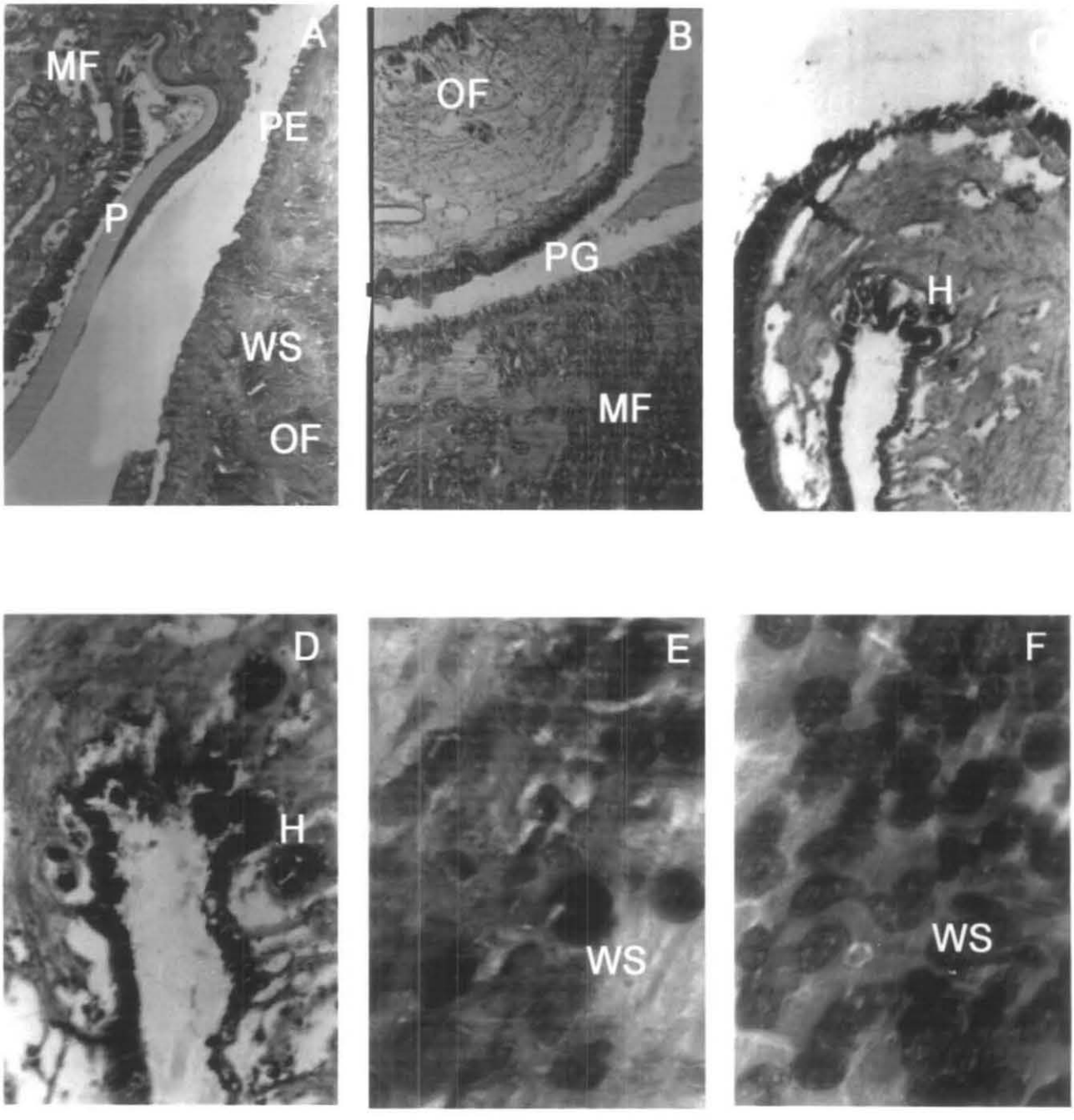


Plate 6. A-F. Light micrographs of mantle tissue of *Perna indica*

layers externally surrounded by epimysium. Fibrous tissues were enclosed in a bundle of muscle fibres which in turn was enclosed by a thin layer of fibrous tissue called endomysium (Plate 7 A & B). Myodegenerative changes marked by hyaline change ie. loss of cross striations and fragmentation of muscle fibres was observed in the adductor muscle of sponge-infested mussel (Plate 7 C & D).

Electron micrographs of cytoplasm of the mantle tissue of normal mussel contained the following organelles; nucleus with prominent chromatin granules (Plate 8 A & C), endoplasmic reticulum, (Plate 8 C & D), endocytic canals and mitochondria (Plate 8 B).

Extensive pathological changes occurred in the mantle tissue of *P. indica* as a result of infestation by boring sponges. Initially, after the periostracum was slit by sponge boring, increased secretory activity occurred in the cells of mantle. The lysosomes underwent a sequence of changes which are reported herein. During the initial stages of shell damage increased activity of lysosomes was observed in the infested mantle cells. As infestation proceeds, cellular autophagy increased and the intensity of autophagy depends on the extent of sponge infestation. The organelles like nucleus, mitochondria and endoplasmic reticulum underwent rapid degradation possibly through the action of lysosomal enzymes leading to cellular dystrophy. The most conspicuous structures present in the cytoplasm of the infested tissue were the electron dense bodies (Plate 10 B, C & D).

Cytopathologic changes in the outer mantle tissue cells of sponge infested brown mussel were hypertrophy of the infested nucleus, loss of chromatin; (Plate 9 D) proliferation and hypertrophy of rough and smooth endoplasmic reticulum in large numbers of cells; (Plate 9 B &10 A) and formation of small vesicles in nucleoplasm of degenerating nuclei of cells (Plate 9 C). Whorl formation (Plate 9 A) and engulfing process of mitochondria (Plate 9 B) were also observed in the infested tissues subjected to shell damage.

Plate 7. A - D

A. Uninfested adductor muscle of brown mussel (C.S, H & E, ×200)

B. Adductor muscle of brown mussel infested by sponge showing myodegeneration (C.S, H & E, ×200)

C. Uninfested adductor muscle of brown mussel (striated portion) (C.S, H &E, ×200)

D. Adductor muscle of brown mussel infested by sponge showing fragmentation of muscle fibre (striated portion) (C.S, H & E, ×200)

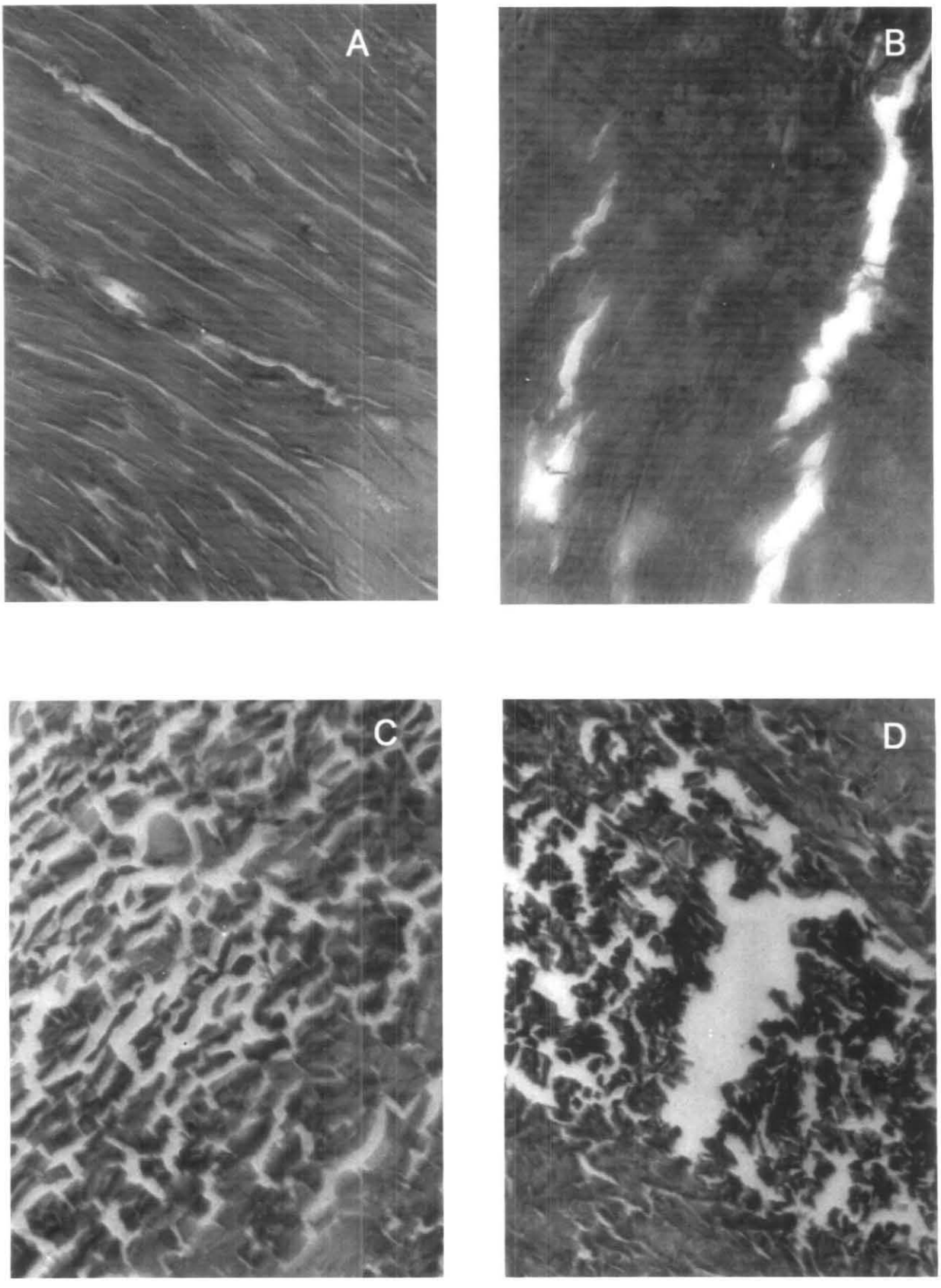


Plate 7. A-D. Light micrographs of uninfested and sponge infested adductor muscle of *Perna indica*

Plate 8. A - D

A. Nucleus (N)× 20,000

B. Endocytic canals (EC), Mitochondria (M) × 40,000

C. Nucleus (N), Endoplasmic reticulum (ER) × 40,000

D. Endoplasmic reticulum (ER) × 50,000

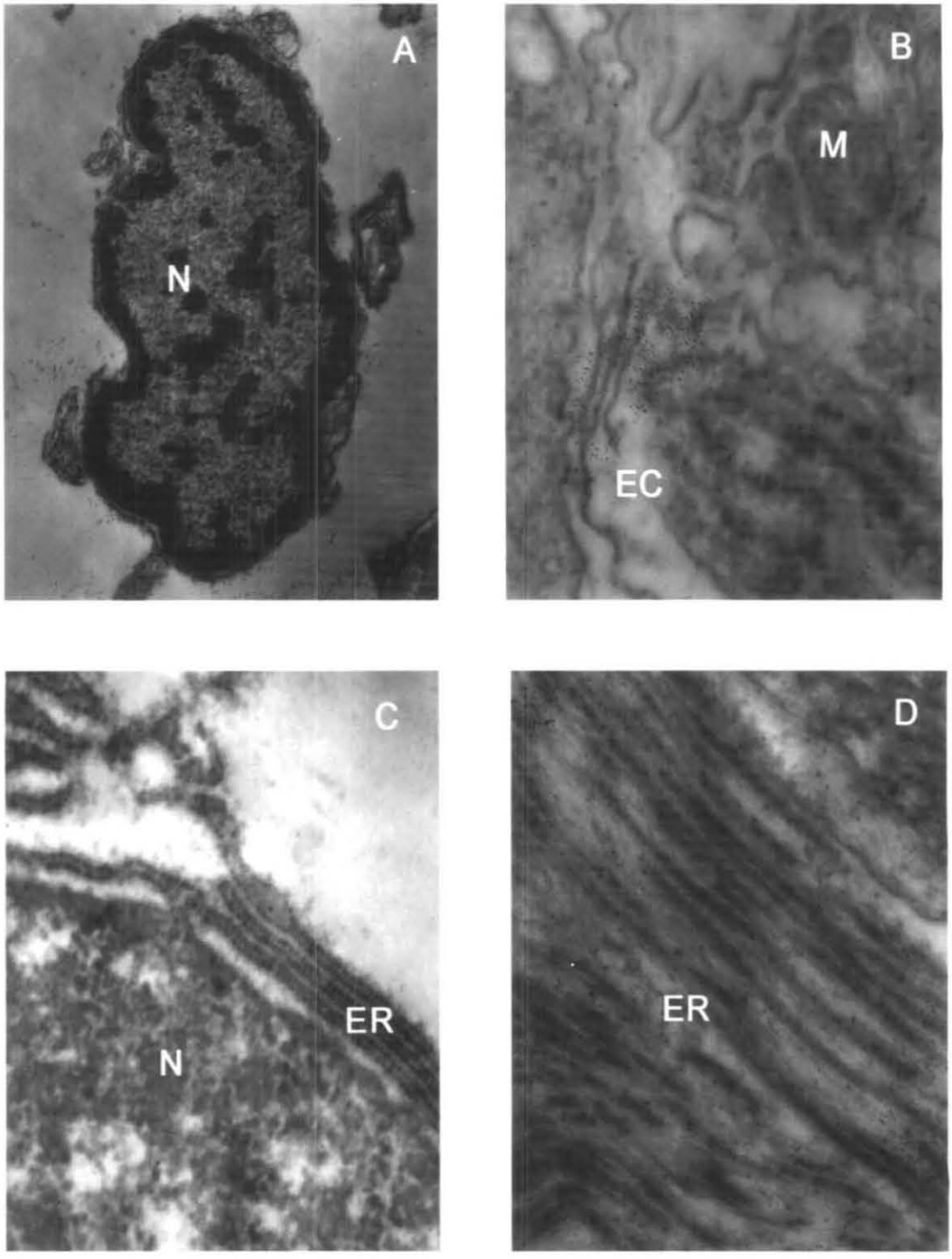


Plate 8 A-D. Electron micrographs of uninfested mantle tissue of *Perna indica*

Plate 9. A - D

A. Whorl formation (W) × 20,000

B. Engulfing mitochondria (M), multivesicular body (MVB) × 40,000

C. Golgi vesicles (GV) × 40,000

D. Degenerating nucleus (N) × 25,000

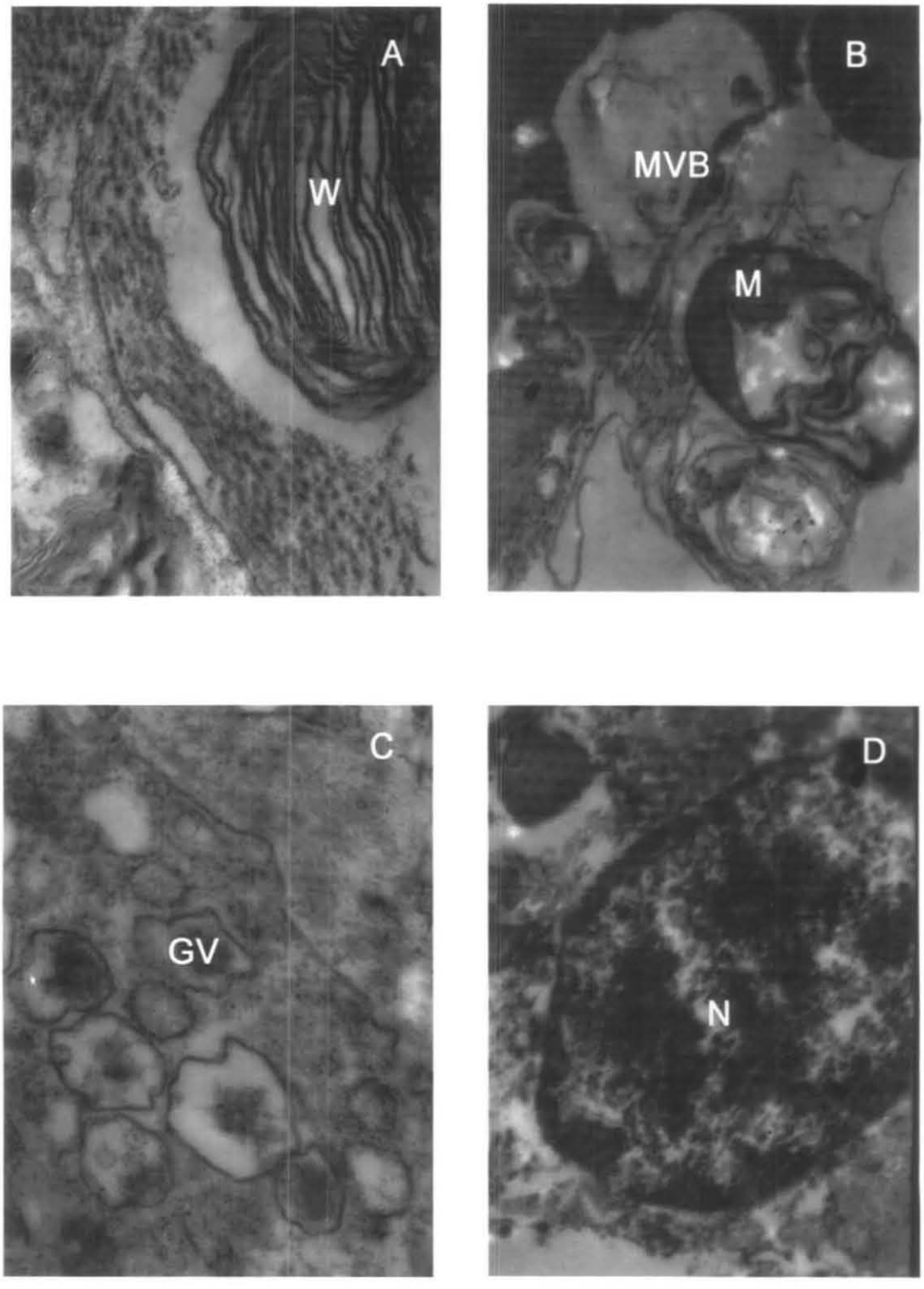


Plate 9. A-D. Electron micrographs of sponge infested mantle tissue of *Perna indica*

Plate 10. A - D

A. Fragmented endoplasmic reticulum (ER), lysosomes (L) × 50,000

B. Cellular dystrophy showing the presence of only electron dense bodies (EB) × 40,000

C. Electron dense bodies migrating towards the peripheral region of cell (EB) × 40,000 **D.** Lysosomes, magnified view of electron dense bodies with granules (L) × 50,000

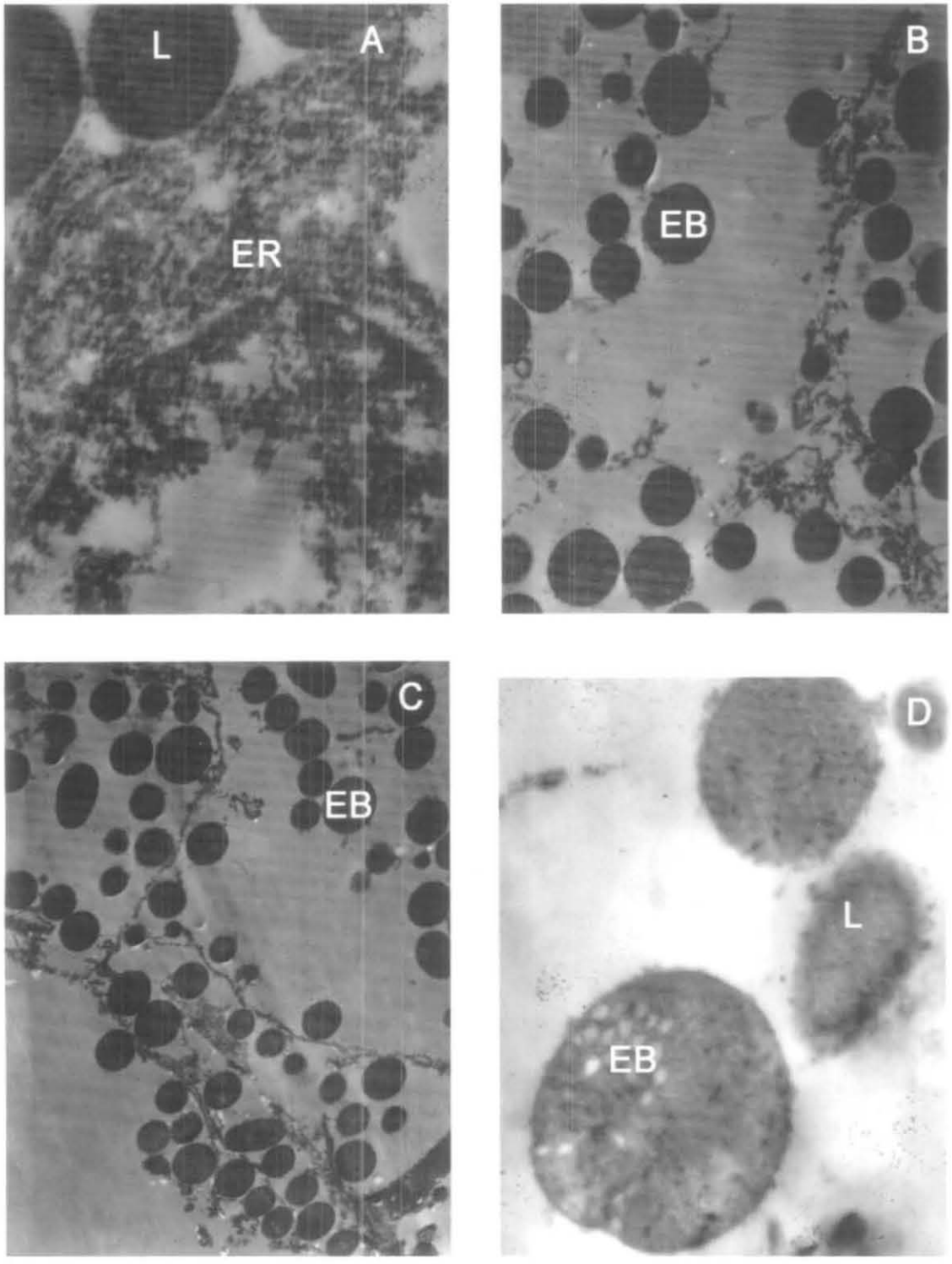


Plate 10. A-D. Electron micrographs of sponge infested mantle tissue of *Perna indica*

5. DISCUSSION

The mantle of molluscs is the primary organ responsible for the formation of shell. The periostracum, the outermost uncalcified coating of molluscan shell, the primary function of which is the deposition of inorganic phase to the shell, is secreted by the mantle at the growing edge of the shell.

When the additional secretion of nacreous material is delayed due to adverse environmental conditions, the sponge makes direct contact with the mantle producing lysis or atrophy of the mantle epithelium; dark pigmented pustules are formed at this spot.

Studies on histology have been useful in assessing the effect of parasites and borers in molluscan tissues. The main cellular response to shell damage is the infiltration of haemocytes into the mantle connective tissue lying adjacent to the epithelial cells underlying the region of calcareous material. Convergence of haemocytes in some areas of infestation indicated that inflammatory reaction might develop by the cells against the infecting agent or the intruder.

Haemocytosis is a typical response of bivalves to parasitisation. Haemocytic infiltration of connective tissue was observed as a major sign of infestation by cytozoic parasite on the oyster *Ostrea edulis* (Balouet *et al.*, 1983). The histopathological progression of haemic neoplasms in bay mussels showed cellular disorders in the connective tissues of mantle which are characterised by large, multiple and pleomorphic nuclei (Mix, 1983). Heavy riddling at or near adductor attachment zone may also cause defective functioning of shell valves (" insertion scar imbalance", Thomas, 1983).

The mantle areas, which normally secrete nacre, may secrete periostracum under abnormal conditions like disease or shell damage. Under these circumstances the mantle shows remarkable changes like development of elongated

cells, infiltration of haemocytes etc. (Dix, 1973). Four different secretory cells were observed in the mantle region of the pearl oyster, *Pinctada mazatlantica*. They are large basophilic secretory cells of epithelial mantle, small secretory cells found in the middle fold, acidophilic secretory cells found in the shell epithelium and secretory cells found in the central zone associated with glycogen synthesis (Garcia-Gasca *et al.*, 1994). The periostracum forms a thin sheet that extends out over the inner surface of the outer lobe that is covered with secretory granules (Morrison, 1993). Prokaryotic infestation in the sea scallop, *Placopecten magellanicus*, resulted in grayish, flaccid adductor muscles with prominent myodegeneration (Gulka and Chang, 1983).

It is possible that accumulation of intra cellular secretions is the result of increased cellular activity induced by stressors. No marked pathological change was observed in the mantle tissue of the gastropod *Chicoreus ramosus* infested by sporocysts of trematodes and nematodes (Kagoo and Ayyakkannu, 1994). The infested animals had a heavily damaged or bored shells. Certain organisms which are capable of boring through the shell may pave way for the entry of secondary parasites, thereby increasing the infestation by parasites. The results of the present study indicated the changes at histological level in the mantle tissue of the host mussel. Even though infestation is restricted mainly to the shell layers, in the case of severe infestation histological changes are resulted in the mantle tissue underlying the shell layers.

The cells of mantle epithelium have shown structural changes and degeneration involving atrophy of cells. Cellular dystrophy and increased secretory activity were observed in the organelles immediately after periostracum damage. Bubel (1973) stated that periostracum repair in damaged cells is similar to that in normal cells. The ultrastructure of the neopilinid mantle edge differs from that of other bivalves in several aspects. The periostracum groove is in fact situated between the inner surface of the median mantle fold and the weakly developed inner mantle fold, a condition not found in any other molluscan group (Schaffer and Haszprunar, 1997). The structure of the molluscan shell is complex, and the ways in which it can be modified are numerous. Basically it consists of an outer covering, the periostracum which is composed of

organic chemicals, and several inner layers of calcium carbonate. Although similar in appearance to the 'chitin' of an arthropod shell, the periostracum is chemically distinct and is usually referred to as 'conchin' or 'conchiolin'. This provides some protection to the underlying calcareous shell layers against erosion or boring organisms (Anandakumar and Ayyakkannu, 1991).

Pustule disease caused a wide variety of pathological changes in abalones. The intense inflammatory response in the centers of the lesions. The lesions first formed on the surface of the abalone foot and tended to migrate into the tissue as the disease progressed (Lee *et al.*, 1997). The disease in advanced stages leads to nuclear membrane disintegration resulting in nucleoplasm diffusion, loss of the nucleolus and mitochondria denaturation. Ultrastructure studies on the mantle tissue of the pearl oyster *Pinctada margaritifera* revealed the presence of distinct epithelial areas: the inner pallial zone made of columnar epithelium, the middle fold made of cuboidal or ciliated cells, the periostracal groove consisted of pseudostratified epithelium, the outer fold of transitional epithelium, the columnar epithelium and the outer pallial zone of cuboidal epithelium (Zahab *et al.*, 1992). The present study on the ultrastructure of mantle epithelium also revealed a similar structural pattern in *Perna indica*.

Specialized basal cells are present at the base of the periostracal groove in bivalves. The pellicle, which is derived from the basal cell, is modified for various adaptations in them. Immediately after shell damage, the cells undergo an increase in secretory activity, ie. increase in the number of lysosomes and multivesicular bodies leading to the formation of an organised layer called proto-ostracum (Bubel, 1973 A). In the present study the electron dense bodies are noticed as a unique feature of sponge infested mantle tissue.

Infestation causes an alteration in haemolymph components, a reduction in growth rate and weakness with respect to valve closure and attachment to the substrate. The development of shellfish-based industries and the concomitant increase in demand for the introduction and transfer of different shellfish species and stocks have

increased the risks of spreading their parasites and diseases around the world. To avoid the accidental introduction of infectious disease agents, information on known parasites and diseases must be readily available. Since the number of recognized infectious agents and facts on known diseases is continuously increasing, it is necessary to update the current state of knowledge on their geographic distribution, host species infested (both naturally and experimentally), impact on host health, diagnostic techniques for many of the diseases and known methods of control.

SUMMARY

With a view to studying the boring sponges infesting the commercially important species of brown mussel, *Perna indica*, Kuriakose and Nair of the southwest coast of India, the present study was initiated at Vizhinjam in 1998 as part of the Ph. D programme in Fish and Fisheries Science (Mariculture) of the Central Institute of Fisheries Education, Mumbai. Considering the extent and magnitude of the brown mussel fishery along the southwest coast from Kovalam to Kanyakumari (Cape Comorin), six centres were selected and data collected for two fishing seasons (ie. October, 1998 to March, 1999 and October 1999 to March, 2000) form the basis of the present thesis entitled "Boring sponge infestation on the mussel *Perna indica*, Kuriakose and Nair, from the southwest coast of India". The salient findings emerged during the above study are presented in the thesis in six chapters.

Chapter I

How shells bored by sponge could be differentiated from others bored by any other boring organisms, say boring mollusc, polychaete, sipunculid etc. is delt with in this chapter. Soon after the settlement of boring sponge larva on the surface of shell it penetrates into the shell forming an initial chamber inside. From the sponge "mass" inside the "chamber" branches are formed and each branch, after a short distance, forms another chamber. Thus the boring sponge proliferates inside the shell in a pattern rather unique for sponges and the same may be called " chamber and canal pattern". The sponge "mass" inside the chamber takes water from outside through minute papillae produced from the "mass" which pierce the outer and inner surfaces of the shell. So any shell bored by sponge may have minute openings at the surface usually more crowded at the umbo part but rather sparse and reticulate in thinner parts of the shell. Such openings are not produced by any other boring organism on the surface of shell.

Chapter II

Sponge is a soft bodied and ill organized animal without organs and organ systems, but how such an organism bores into hard calcareous shell/ coral is a matter of puzzle. In the process of boring, sponge chips off minute calcareous particles (also called microchips) of uniform size and shape (mean size of chip: 56× 47×32 μm) from the interior of the shell/ coral and are expelled out through the excurrent stream of water incessantly. A small concavity (diameter: 0.04 to 0.09 mm) is formed at the inner side of the cavities found inside the shell and hence the inner surfaces of chambers and canals made inside the shell may have a frothy appearance under high magnification. This character is unique for boring sponges. As microchips are removed from the substratum (shell) the total weight of the shell falls considerably and the interior becomes more and more cavernous and the shell fragile. Since small particles are removed from the interior this phenomenon may even be called "bioerosion at microlevel or microerosion". Microerosion of coral reefs, hence, is a matter of geological importance as the calcareous frame work of coral reef becomes weak by excessive boring.

Sponges chip off minute "microchips" by means of enzymatic action. Some specialized cells of archaeocytic origin present at the site of contact with the shell are responsible for the etching of these chips. Filopodia formed from these cells form fine cervices around a future chip and by progressive stages of plasmolysis the chip is cut and removed from its original position by filopodial basket developed around a "future chip". When a microchip is thus scooped out from the interior of any cavity formed inside the shell by the sponge a concavity (pit) is formed. Details of chips, pits and their measurements are given in this chapter with reference to mussel shell.

Chapter III

The taxonomy of ten species of boring sponges occurring in the southwest coast is delt with in detail in this chapter. Out of a total of 10 species, 9 are seen as pests of brown mussel while one as pest of rock oyster. For the comprehensiveness of the account all the ten species are included in the list and a key is also provided for their identification.

A restricted synonymy is provided for each species and the description includes details of galleries formed inside the shell, pathological aspects, spicular details and the zoogeography.

The following new distributional records have also been made during the present study. *Alectona millari* was here recorded from the Indian Ocean (Locality: Enayam); both *Thoosa hancocki* and *T. armata* were here recorded as pests of brown mussel; *Aka minuta* could be recorded as pest of pearl oyster at Tuticorin and *Halina extensa* was here confirmed as a boring species.

Chapter IV

A historical account of the boring sponge investigations, in the Indian context, is presented as an introduction to this chapter and is followed by the investigations carried out during the present study.

In order to get a synoptic picture of boring sponge infestation in brown mussel six centres were selected and samples were collected statistically. Since mussel fishing is seasonal year round collection was not possible and hence the sampling was restricted to two seasons.

From samples thus collected the size frequency distribution of bored and unbored specimens were prepared and the incidence (infested shells/ 100) and species composition (percentage of each boring species/ total infested shells) were calculated monthly and seasonally for each centre.

Statistical analyses of data collected revealed that sponge infestation starts in mussel when they attain a size of 40-45 mm and the intensity of infestation increases as they become older and older.

There was no sponge infestation on brown mussel collected from Kanyakumari; but the same noted at Enayam was the highest, 63.16 % and 42.6 % respectively during the first and second seasons. All other centres recorded lower incidence.

Two unconventional boring species (*Cliona lobata* and *C. margaritifera*) appeared at Vizhinjam around 1980 and started spreading to all natural molluscan beds in and around Vizhinjam. These two new invaders, which were quite notorious for their devastating capacities elsewhere, have effected a sudden hike in the incidence and species composition of boring sponges in every bed. They competed with all conventional species of boring sponges in all beds and suppressed some initially activating others occasionally. In the competition between *C. lobata* and *C. margaritifera* (two new invaders) *C. lobata* dominated in almost all beds and suppression of *C. margaritifera* activated the spreading of *C. vastifica*, a conventional species. Such suppression of *C. margaritifera* indirectly helped to bring down the incidence to a lower level. The lower incidence noticed in the various beds at present may be due to this peculiar interaction. But at Enayam all the above 3 species are represented with equal vigor and this may be the reason for a higher incidence seen at Enayam.

The present study, made after a period of 20 years from the date of first entry of boring sponges to Vizhinjam culture rafts, hence, may be taken as a follow-up on their distribution and abundance in the Indian beds during this interim period.

Other aspects presented in this chapter include month-wise and season-wise incidence, biodiversity. Maximum number of boring sponge species was recorded at Enayam (8 nos.) while in all other centres their number fluctuated between 4 and 5.

Chapter V

Migration of boring sponges from the wild to tended stocks is delt with in this chapter based on primary data collected from Tuticorin pearl oyster and Vizhinjam mussel culture rafts. Attempts were made to culture brown mussel in the Ashtamudi Lake, Kollam, to trace out the pattern of migration of boring sponges to a cultivated stock in an estuary.

At Vizhinjam, *C. lobata* (new invader) and *C. vastifica* (conventional species) occupied the first two ranks respectively (on rafts) while *C. margaritifera* occupied the third position. The incidence was found to be 23 % which is considerably high when compared to that in the wild stocks off Vizhinjam.

The boring sponge fauna of Ashtamudi Lake comprised of one euryhaline species, *C. vastifica*, with an incidence of 18 %.

Chapter IV

The changes which take place inside the shell and also in the soft tissue of the infested mussel are discussed in this chapter. Of some 12 different diseases reported from the molluscan shells to-date, only 6 diseases could be detected during the present study. Of these blister formation is quite wide spread followed by discoloration of mantle tissue and porosis (bifacial).

TEM studies made on soft tissue revealed an array of pathological manifestations. Distinct pathological symptoms like increased secretion of wandering secretory cells, haemocytosis, vacuolisation of mantle epithelial cells, sloughening of upper epithelial layer, increased activity of lysosomes, rapid degeneration of organelles like nucleus, mitochondria etc, were evident in the host. Cytoplasm of infested tissue was loaded with "electron dense bodes'. In adductor muscle myodegradative changes, including fragmentation of muscle fibres, were common and this evidently affected the opening and closing of the shell valves of the live mussel.

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